

# NO SURE FIX

Prospects for Reducing Nitrogen Fertilizer  
Pollution through Genetic Engineering



Union of Concerned Scientists  
Citizens and Scientists for Environmental Solutions



# NO SURE FIX

Prospects for Reducing Nitrogen Fertilizer Pollution  
through Genetic Engineering

Doug Gurian-Sherman

Noel Gurwick

*Union of Concerned Scientists*

*December 2009*

© 2009 Union of Concerned Scientists  
All rights reserved

**Doug Gurian-Sherman** and **Noel Gurwick** are senior scientists in the Union of Concerned Scientists (UCS) Food and Environment Program.

The Union of Concerned Scientists (UCS) is the leading science-based nonprofit working for a healthy environment and a safer world. UCS combines independent scientific research and citizen action to develop innovative, practical solutions and to secure responsible changes in government policy, corporate practices, and consumer choices.

The goal of the UCS Food and Environment Program is a food system that encourages innovative and environmentally sustainable ways to produce high-quality, safe, and affordable food, while ensuring that citizens have a voice in how their food is grown.

More information is available on the UCS website at [www.ucsusa.org/food\\_and\\_agriculture](http://www.ucsusa.org/food_and_agriculture).

This report is available on the UCS website (in PDF format) at [www.ucsusa.org/publications](http://www.ucsusa.org/publications) or may be obtained from:

UCS Publications  
2 Brattle Square  
Cambridge, MA 02238-9105

Or, email [pubs@ucsusa.org](mailto:pubs@ucsusa.org) or call (617) 547-5552.

DESIGN: Catalano Design

COVER IMAGE: Todd Andraski/University of Wisconsin-Extension

# Contents

---

TEXT BOXES, FIGURES, AND TABLES	<i>iv</i>
ACKNOWLEDGMENTS	<i>v</i>
EXECUTIVE SUMMARY	1
<b>CHAPTER 1: INTRODUCTION: GENETIC ENGINEERING AND NITROGEN IN AGRICULTURE</b>	5
Key Terms Used in This Report	5
Report Organization	6
The Impact of Nitrogen Fertilizer Use in Agriculture	6
The Role of Reactive Nitrogen	8
<b>CHAPTER 2: NITROGEN USE EFFICIENCY IN GE PLANTS AND CROPS</b>	10
How We Evaluated GE's Prospects for Improving NUE	11
Studies of GE NUE Crops	12
Approved Field Trials of GE NUE Crops	15
Possible Risks Related to GE NUE Genes	16
Commercialization of GE NUE Crops	17
<b>CHAPTER 3: IMPROVING NUE THROUGH TRADITIONAL AND ENHANCED BREEDING METHODS</b>	19
NUE Improvements in Commercial Varieties	19
The Impact of Higher Yield on NUE	19
Genetic Variability of NUE-Related Traits in Major Crops	20
Strengths and Limitations of Breeding Compared with GE	22
<b>CHAPTER 4: THE ECOSYSTEM APPROACH TO NUE</b>	24
A Big-Picture Perspective	24
The Time Is Ripe for a New Approach	24
<b>CHAPTER 5: OTHER MEANS OF IMPROVING NUE</b>	26
Cover Crops	27
Precision Farming	28
<b>CHAPTER 6: CONCLUSIONS AND RECOMMENDATIONS</b>	30
The Promise and Pitfalls of Non-GE Approaches	30
What the United States Should Do	31
REFERENCES	33

## *Text Boxes, Figures, and Tables*

---

### **TEXT BOXES**

- |  |    |
|--|----|
| 1. How Engineered Genes Contribute to Plant Traits | 10 |
| 2. Methods Used to Study Crop NUE                  | 11 |

### **FIGURES**

- |   |    |
|---|----|
| 1. The Nitrogen Cycle                     | 7  |
| 2. Rise in Reactive Nitrogen Production   | 8  |
| 3. USDA-Approved Field Trials of GE Crops | 15 |

### **TABLES**

- |  |    |
|--|----|
| 1. Genes Used to Improve NUE through Genetic Engineering | 13 |
| 2. Improvements in Nitrogen Use Efficiency               | 20 |

## *Acknowledgments*

---

This report was made possible in part through the generous financial support of C.S. Fund, Clif Bar Family Foundation, CornerStone Campaign, Deer Creek Foundation, The Educational Foundation of America, The David B. Gold Foundation, The John Merck Fund, Newman's Own Foundation, Next Door Fund of the Boston Foundation, The David and Lucile Packard Foundation, and UCS members.

The authors would like to thank Walter Goldstein of the Michael Fields Agricultural Research Institute, Linda Pollack of the U.S. Department of Agriculture's Agricultural Research Service, and Christina Tonitto of Cornell University. The time they spent in reviewing this report is greatly appreciated and significantly enhanced the final product.

Here at UCS, the invaluable insights provided by Mardi Mellon and Karen Perry Stillerman helped clarify and strengthen the report as well. Brenda Ekwurzel contributed valuable suggestions regarding climate-change-related aspects of the report. The authors also thank Heather Sisan for research assistance that made everything go more smoothly. Finally, the report was made more readable by the expert copyediting of Bryan Wadsworth.

The opinions expressed in this report do not necessarily reflect the opinions of the foundations that support the work, or the individuals who reviewed and commented on it. Both the opinions and the information contained herein are the sole responsibility of the authors.



## Executive Summary

---

**N**itrogen is essential for life. It is the most common element in Earth's atmosphere and a primary component of crucial biological molecules, including proteins and nucleic acids such as DNA and RNA—the building blocks of life.

Crops need large amounts of nitrogen in order to thrive and grow, but only certain chemical forms collectively referred to as reactive nitrogen can be readily used by most organisms, including crops. And because soils frequently do not contain enough reactive nitrogen (especially ammonia and nitrate) to attain maximum productivity, many farmers add substantial quantities to their soils, often in the form of chemical fertilizer.

Unfortunately, this added nitrogen is a major source of global pollution. Current agricultural practices aimed at producing high crop yields often result in excess reactive nitrogen because of the difficulty in matching fertilizer application rates and timing to the needs of a given crop. The excess reactive nitrogen, which is mobile in air and water, can escape from the farm and enter the global nitrogen cycle—a complex web in which nitrogen is exchanged between organisms and the physical environment—becoming one of the world's major sources of water and air pollution.

The challenge facing farmers and farm policy makers is therefore to attain a level of crop productivity high enough to feed a growing world population while reducing the enormous impact of nitrogen pollution. Crop genetic engineering has been proposed as a means of reducing the loss of reactive nitrogen from agriculture. This report represents a first step in evaluating the prospects of genetic engineering to achieve this goal while increasing crop productivity, in comparison with

other methods such as traditional crop breeding, precision farming, and the use of cover crops that supply reactive nitrogen to the soil naturally.

### The Importance of Nitrogen Use Efficiency (NUE)

Crops vary in their ability to absorb nitrogen, but none absorb all of the nitrogen supplied to them. The degree to which crops utilize nitrogen is called nitrogen use efficiency (NUE), which can be measured in the form of crop yield per unit of added nitrogen. NUE is affected by how much nitrogen is added as fertilizer, since excess added nitrogen results in lower NUE. Some agricultural practices are aimed at optimizing the nitrogen applied to match the needs of the crop; other practices, such as planting cover crops, can actually remove excess reactive nitrogen from the soil.

In the United States, where large volumes of chemical fertilizers are used, NUE is typically below 50 percent for corn and other major crops—in other words, more than half of all added reactive nitrogen is lost from farms. This lost nitrogen is the largest contributor to the “dead zone” in the Gulf of Mexico—an area the size of Connecticut and Delaware combined, in which excess nutrients have caused microbial populations to boom, robbing the water of oxygen needed by fish and shellfish. Furthermore, nitrogen in the form of nitrate seeps into drinking water, where it can become a health risk (especially to pregnant women and children), and nitrogen entering the air as ammonia contributes to smog and respiratory disease as well as to acid rain that damages forests and other habitats. Agriculture is also the largest human-caused domestic source of nitrous oxide, another reactive form of nitrogen that contributes to global

warming and reduces the stratospheric ozone that protects us from ultraviolet radiation.

Nitrogen is therefore a key threat to our global environment. A recent scientific assessment of nine global environmental challenges that may make the earth unfavorable for continued human development identified nitrogen pollution as one of only three—along with climate change and loss of biodiversity—that have already crossed a boundary that could result in disastrous consequences if not corrected. One important strategy for avoiding this outcome is to improve crop NUE, thereby reducing pollution from reactive nitrogen.

### **Can Genetic Engineering Increase NUE?**

Genetic engineering (GE) is the laboratory-based insertion of genes into the genetic material of organisms that may be unrelated to the source of the genes. Several genes involved in nitrogen metabolism in plants are currently being used in GE crops in an attempt to improve NUE. Our study of these efforts found that:

- Approval has been given for approximately 125 field trials of NUE GE crops in the United States (primarily corn, soybeans, and canola), mostly in the last 10 years. This compares with several thousand field trials each for insect resistance and herbicide tolerance.
- About half a dozen genes (or variants of these genes) appear to be of primary interest. The exact number of NUE genes is impossible to determine because the genes under consideration by companies are often not revealed to the public.
- No GE NUE crop has been approved by regulatory agencies in any country or commercialized, although at least one gene (and probably more) has been in field trials for about eight years.
- Improvements in NUE for experimental GE crops, mostly in controlled environments, have typically ranged from about 10 to 50 percent for grain crops, with some higher values.

There have been few reports of values from the field, which may differ considerably from lab-based performance.

- By comparison, improvement of corn NUE through currently available methods has been estimated at roughly 36 percent over the past few decades in the United States. Japan has improved rice NUE by an estimated 32 percent and the United Kingdom has improved cereal grain NUE by 23 percent.
- Similarly, estimates for wheat from France show an NUE increase from traditional breeding of about 29 percent over 35 years, and Mexico has improved wheat NUE by about 42 percent over 35 years.

Available information about the crops and genes in development for improved NUE suggests that these genes interact with plant genes in complex ways, such that a single engineered NUE gene may affect the function of many other genes. For example:

- In one of the most advanced GE NUE crops, the function of several unrelated genes that help protect the plant against disease has been reduced.
- Another NUE gene unexpectedly altered the output of tobacco genes that could change the plant's toxicological properties.

Many unexpected changes in the function of plant genes will not prove harmful, but some may make it difficult for the crops to gain regulatory approval due to potential harm to the environment or human health, or may present agricultural drawbacks even if they improve NUE. For the most advanced of the genes in the research pipeline, commercialization will probably not occur until at least 2012, and it will likely take longer for most of these genes to achieve commercialization—if they prove effective at improving NUE. At this point, the prospects for GE contributing substantially to improved NUE are uncertain.

## Other Methods for Reducing Nitrogen Pollution

*Traditional or enhanced breeding techniques* can use many of the same or similar genes that are being used in GE, and these methods are likely to be as quick, or quicker, than GE in many cases. Traditional breeding may have advantages in combining several NUE genes at once.

*Precision farming*—the careful matching of nitrogen supply to crop needs over the course of the growing season—has shown the ability to increase NUE in experimental trials. Some of these practices are already improving NUE, but adoption of some of the more technologically sophisticated and precise methods has been slow.

*Cover crops* are planted to cover and protect the soil during those months when a cash crop such as corn is not growing, often as a component of an organic or similar farming system. Some can supply nitrogen to crops in lieu of synthetic fertilizers, and can remove excess nitrogen from the soil; in several studies, cover crops reduced nitrogen losses into groundwater by about 40 to 70 percent.

Cover crops and other “low-external-input” methods (i.e., those that limit use of synthetic fertilizers and pesticides) may also offer other benefits such as improving soil water retention (and drought tolerance) and increasing soil organic matter. An increase in organic matter that contains nitrogen can reduce the need for externally supplied nitrogen over time.

With the help of increased public investment, these methods should be developed and evaluated fully, using an ecosystem approach that is best suited to determine how reactive nitrogen is lost from the farm and how NUE can be improved in a comprehensive way. Crop breeding or GE alone is not sufficient because they do not fully address the nitrogen cycle on real farms, where nitrogen loss varies over time and space, such as those times when crops—conventional or GE—are not growing.

## Conclusions

GE crops now being developed for NUE may eventually enter the marketplace, but such crops

are not uniquely beneficial or easy to produce.

There is already sufficient genetic variety for NUE traits in crops, and probably in close relatives of important crops, for traditional breeding to build on its successful track record and develop more efficient varieties.

Other methods such as the use of cover crops and precision farming can also improve NUE and reduce nitrogen pollution substantially.

## Recommendations

The challenge of optimizing nitrogen use in a hungry world is far too important to rely on any one approach or technology as a solution. We therefore recommend that research on improving crop NUE continue. For traditional breeding to succeed, public research support is essential and should be increased in proportion to this method’s substantial potential.

We also recommend that system-based approaches to increasing NUE—cover crops, precision application of fertilizer, and organic or similar farming methods—should be vigorously pursued and supported. These approaches are complementary to crop improvement because each addresses a different aspect of nitrogen use. For example, while breeding for NUE reduces the amount of nitrogen needed by crops, precision farming reduces the amount of nitrogen applied. Cover crops remove excess nitrogen and may supply nitrogen to cash crops in a more manageable form.

Along with adequate public funding, incentives that lead farmers to adopt these practices are also needed. Although the private sector does explore traditional breeding along with its heavy investment in the development of GE crops, it is not likely to provide adequate support for the development of non-GE varieties, crops that can better use nitrogen from organic sources, or improved cover crops that remove excess nitrogen from soil. We must ensure that broad societal goals are addressed and important options are pursued nevertheless.

In short, there are considerable opportunities to address the problems caused by our current

overuse of synthetic nitrogen in agriculture if we are willing to make the necessary investments. The global impact of excess reactive nitrogen will worsen as our need to produce more food increases, so strong actions—including significant investments in technologies and methods now largely ignored by industrial agriculture—will be required to lessen the impact.

## CHAPTER 1

# *Introduction: Genetic Engineering and Nitrogen in Agriculture*

---

**T**he need to raise global food production perhaps as much as 100 percent by the middle of the century poses one of the major challenges currently facing the world—as does reducing the pollution caused by many current agricultural practices. Because plant growth is often constrained by the amount of nitrogen in the soil that plants can access, adding more nitrogen to agricultural fields will almost certainly play a role in meeting the challenge of increased crop productivity. Unfortunately, some of the nitrogen sources readily available to farmers across much of the globe are already chief contributors to nitrogen pollution.

Dobermann and Cassman (2005) project a need to increase grain production 38 percent by 2025, and assert that this may be done with a nitrogen crop yield response increase of 20 percent using current technologies, with a net increase in nitrogen of 30 percent if current losses of agricultural land do not continue. Other estimates, however, note that a 45 percent reduction in nitrogen pollution in the Gulf of Mexico is likely needed to have a substantial impact on the dead zone there (EPA 2009b). Pouring on even more fertilizer to increase food production would aggravate this and other problems and carry potentially high costs. What we need are ways to increase food production on existing farmland while reducing nitrogen pollution.

Strategies for reducing nitrogen loss from farms without reducing productivity include vegetation buffer strips planted along waterways adjacent to crop fields; such buffers have captured significant

amounts of nitrogen that would otherwise reach streams and rivers. Also, better timing of nitrogen fertilizer application—to be performed only when it is actually needed by a given crop during the growing season—reduces the amount of nitrogen applied.

### **Key Terms Used in This Report**

Improving the *nitrogen use efficiency (NUE)* of crops is another strategy for reducing nitrogen loss from farms—and consequent downstream nitrogen pollution—in this case by increasing the amount of plant growth that occurs for each pound of nitrogen added to the soil. Improved NUE reduces the need for nitrogen fertilizer. This can potentially be done in two ways: through traditional or enhanced methods of crop breeding, or through genetic engineering.

NUE can also be improved in order to reduce nitrogen loss from farm fields rather than to increase crop yield. The use of cover crops and better-timed fertilizer applications often serve this purpose. It should be noted that because different methods for measuring NUE can arrive at different values, it may be difficult to make direct comparisons between NUE values found in this report and elsewhere.

*Traditional breeding* involves controlled mating between plant parents selected for their desirable traits. This powerful technology, responsible for most genetic improvement in crops over the last 100 years, can now be enhanced with new genomic technologies that assist scientists in identifying prospective traits. Using information about plant genetics to inform breeding does not constitute

genetic engineering, and the promise offered by these two approaches may differ dramatically.

*Genetic engineering (GE)* refers specifically to the isolation and removal of genes—specifically, genes that determine traits of scientific or economic interest—from one organism and their insertion into another, where they become part of the inherited genetic material. In relation to crops, GE can add genes to plants from virtually any source and achieve gene combinations not possible in nature. For example, most commercialized GE crops contain genes from bacteria that make the crops immune to certain herbicides or protect them against insect pests.

GE and traditional breeding have different advantages and limitations as techniques for developing new crop varieties. GE enables us to combine genes from organisms that cannot reproduce with each other, but its success depends on how specific genes (and specific combinations of genes) influence plant growth. Very few plant traits are controlled by a single gene, and our understanding of how multi-gene systems influence plant growth is limited, especially when considering the varied environmental conditions under which plants grow and the changes in gene function and metabolism that occur over the life of the plant.

Traditional breeding, which is sometimes informed by a detailed understanding of the parent plants' genetics, also rearranges the genetic material of the crop. But in this case, because all of the parents' genes are involved, some undesired genes may end up in the resulting crop along with the genes of interest. And unlike GE it uses only those genes already found in the crop or closely related plant species. The ability of traditional breeding to bring many genes from sexually compatible plants together can be advantageous for improving the many traits controlled by multiple genes. While knowledge of genetics can inform traditional breeding, this method can also achieve the desired traits even when the genetic basis is not thoroughly understood.

## Report Organization

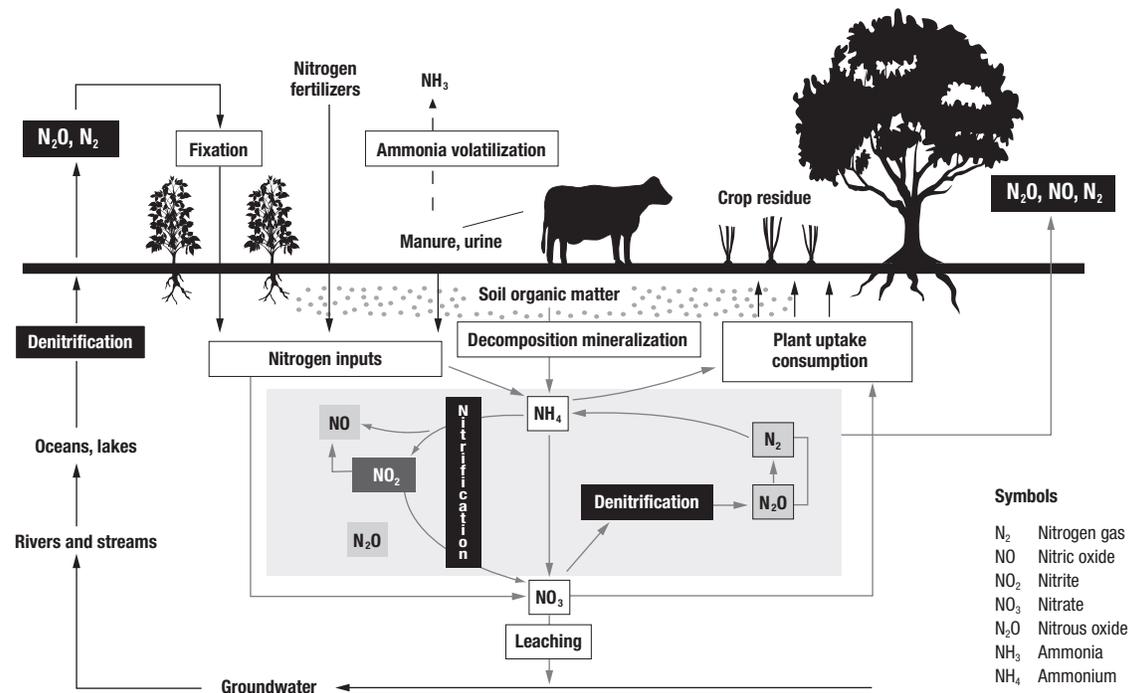
This report describes the status of GE as a tool for producing crops with improved NUE, and is divided along the following lines:

- The next section of Chapter 1 describes the role of the nitrogen cycle.
- Chapter 2 provides definitions for NUE relevant to this report and discusses the implications of using different conceptual frameworks to measure NUE. We then evaluate GE's prospects for providing food and feed crops with enhanced NUE, based on an examination of the scientific literature and government databases.
- Chapter 3 evaluates traditional breeding's prospects for providing food and feed crops with enhanced NUE. Covered technologies include marker-assisted breeding and other advances in genomics, and the identification of crop genes involved in nitrogen metabolism. Important differences between traditional breeding and GE are considered.
- To provide appropriate context, Chapter 4 discusses the value of an ecosystem approach to evaluating nitrogen pollution and solutions, and Chapter 5 reviews two other approaches for reducing fertilizer use and nitrogen pollution: precision farming and cover cropping.
- Finally, Chapter 6 offers several recommendations for public policies that can help reduce nitrogen pollution.

## The Impact of Nitrogen Fertilizer Use in Agriculture

The addition of nitrogen fertilizers, along with other changes in agriculture, has greatly increased crop productivity in many parts of the world, allowing global food production to remain ahead of rapid population growth in the second half of the twentieth century (Vitousek et al. 2009). But areas where soils are exceptionally deficient in nitrogen, such as much of Africa (Sanchez 2002),

Figure 1. The Nitrogen Cycle



*The nitrogen cycle is a highly complex, global cycle that continuously transforms nitrogen into various chemical forms. Industrial agriculture—with its inefficient use of synthetic fertilizers—alters this cycle by adding excessive amounts of reactive nitrogen to the local and global environments.*

Source: Adapted from Government of South Australia, Primary Industries and Resources SA.

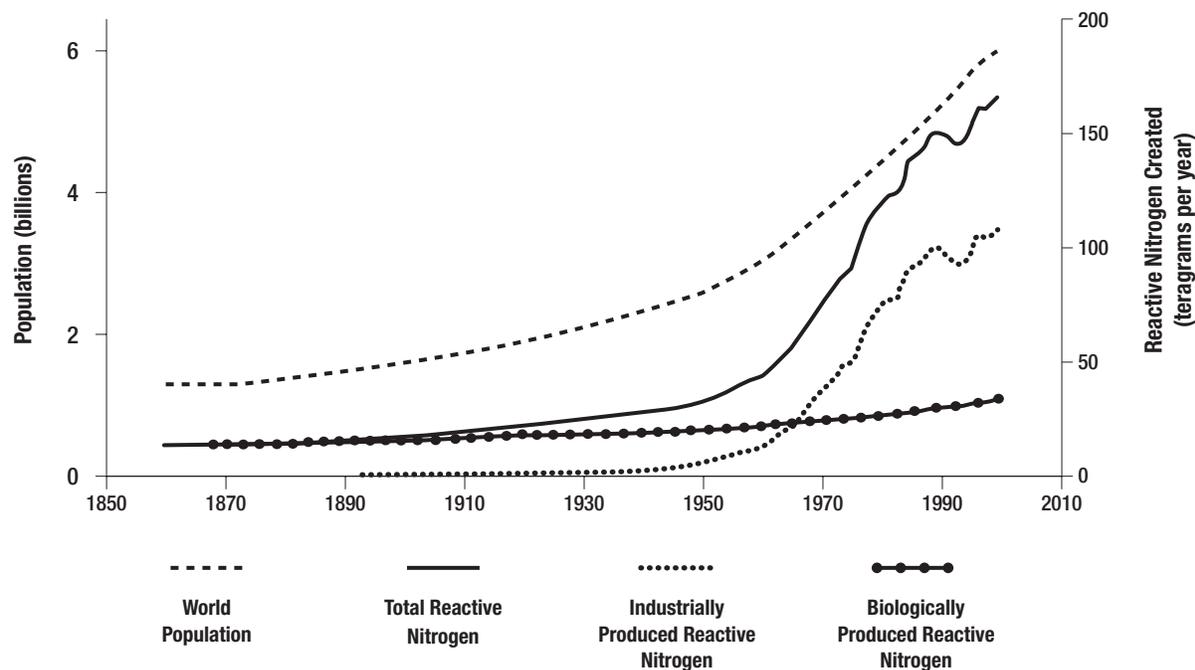
have not kept pace in producing enough food, and improvements in soil fertility are urgently needed.

While essential to food production, nitrogen compounds added to agricultural ecosystems are also some of the most important sources of pollution nationally and globally. Consequences of nitrogen pollution include toxic algal blooms, oxygen-depleted dead zones in coastal waters, and the exacerbation of global climate change, acid rain, and biodiversity loss (Krupa 2003; McCubbin et al. 2002; Vitousek et al. 1997). Reactive nitrogen entering the Mississippi River from crop fields comprises about 42 percent of the nitrogen causing the dead zone in the Gulf of Mexico—at 16,500 sq. km in recent years (EPA 2008), an area the size of Delaware and Connecticut combined.

Fertilizer-intensive agriculture practices are also the United States' major anthropogenic (i.e.,

human-caused) source of nitrous oxide ( $N_2O$ ), a potent heat-trapping gas that also contributes to the destruction of stratospheric ozone. Agricultural soils are responsible for about two-thirds of the anthropogenic nitrous oxide produced in the United States (EPA 2009a). In addition, gaseous ammonia released from nitrogen fertilizer contributes to fine particulate matter that causes respiratory disease and acid rain (Anderson, Strader, and Davidson 2003; Krupa 2003; McCubbin et al. 2002; Vitousek et al. 1997). Nitrate concentrations above 10 parts per million in drinking water have been implicated as a cause of methemoglobinemia, or “blue baby syndrome” (Fan and Steinberg 1996).

Recently, it has been suggested that disruption of the global nitrogen cycle—the complex web in which nitrogen is exchanged between organisms and the physical environment (Figure 1)—caused

**Figure 2. Rise in Reactive Nitrogen Production**

*The amount of human-caused reactive nitrogen in the global environment has increased 11-fold since the nineteenth century and about eight-fold since the 1960s, which marked the beginning of the “green revolution” in agriculture. Agriculture is responsible for about 80 percent of the reactive nitrogen produced worldwide.*

Source: Adapted from Galloway et al. 2003. © 2003, American Institute of Biological Sciences. Used by permission. All rights reserved.

by added nitrogen now exceeds the planet’s capacity to maintain a desirable state for human survival and development (Rockström et al. 2009). Of the nine significant planetary processes or conditions described in that report, only climate change and loss of biodiversity have also passed such a point. This assessment underscores the enormous impact that excess nitrogen is having on the environment.

### The Role of Reactive Nitrogen

The dramatic consequences of nitrogen fertilizer use, both positive and negative, are understandable when we appreciate the extent to which human activity altered the nitrogen cycle in the twentieth century, especially following the “green revolution” of the 1960s (Figure 2). Overall, production of reactive nitrogen increased by a factor of 11, from about

15 teragrams (Tg), or trillion grams, of nitrogen per year in 1860 to about 165 Tg per year in 2000. About 80 percent of this nitrogen has been used in crop production (Galloway et al. 2002).

Those forms of nitrogen called reactive nitrogen are critically important in the context of crop production and its environmental impact. Although nitrogen exists in many forms in the environment and is abundant in the atmosphere as nitrogen gas ( $N_2$ ), this report focuses on two of the many reactive nitrogen compounds most readily used by crops: ammonia and nitrate. These compounds are readily used by both plants and microbes, hence are commonly referred to as reactive nitrogen. By contrast,  $N_2$  cannot be used by most organisms. Reactive nitrogen enters agricultural systems from several sources:

- Industrial production of synthetic fertilizer, which combines natural gas and  $N_2$  to produce ammonia
- Microbe-driven decomposition of organic matter
- Bacterial nitrogen fixation, the process in which microbes, often associated with legumes such as soybeans and alfalfa, break the  $N_2$  bond
- Lightning, which can split the  $N_2$  bond

Agriculture is often the most important source of several reactive nitrogen compounds in the environment. Nitrate, for example, is one of the major forms of reactive nitrogen in fertilizer, and a major source of water pollution. Much of the other major forms of reactive nitrogen in fertilizer, ammonia and urea, are rapidly converted to nitrate. Nitrate is a particular problem because it is especially mobile in the soil, and therefore readily lost through leaching.

The mobility of several forms of reactive nitrogen means that nitrogen can pollute the

environment at local, regional, and global levels. In addition, microbes in soils often convert less mobile forms of reactive nitrogen into more mobile forms such as ammonia and nitrous oxide, which are mobile in the air, further contributing to the spread of nitrogen pollution from farms.

We thus face the dilemma of expanding our food supply to meet the needs of a growing global population—for which we currently rely on increased nitrogen use—while reducing pollution from nitrogen. Whether supplied as synthetic fertilizer or via the addition of biological components like legumes, nitrogen is an expensive input into an agricultural system, so farmers already want to use it as efficiently as possible. But this objective has gained new urgency as we witness the impact of nitrogen overuse on global ecosystems. It is now imperative that we develop new ways of using nitrogen efficiently if we are to avoid even greater harm to the environment in our quest for more food.

## CHAPTER 2

*Nitrogen Use Efficiency in GE Plants and Crops*

The variety of strategies available for increasing NUE (and thereby reducing nitrogen pollution) reflects the different spatial and time scales at which NUE can be analyzed. At the scale of the individual plant, NUE can be increased by enhancing the capacity of that crop species to acquire nitrogen from the soil or better use nitrogen within the plant. For example, a plant with a mature root system that continues to acquire nitrogen even when concentrations in the soil are low—or that acquires nitrogen more rapidly even when concentrations in the soil are high—will use more of the available nitrogen in the soil than a comparable plant with lower NUE.

Similarly, plants that transfer more nitrogen to the grain or increase grain yields will also use nitrogen more efficiently.

Plant characteristics that influence NUE include the amount of energy allocated to root systems (more extensive root systems can enable greater utilization of soil nitrogen) and the specific characteristics of enzyme systems used to acquire nitrogen and allocate acquired nitrogen to different parts of the plant, such as the seed of grain plants. Because the main advantage of GE is its ability to target specific plant traits (Box 1), we here review the status of GE technology for improving NUE, primarily at the scale of the individual plant.

**Box 1. How Engineered Genes Contribute to Plant Traits**

Genes can be thought of as consisting of two parts: the part that carries information needed to produce proteins that underlie traits (the **structural gene**), and the part that directs when and how much of the protein is produced, especially the part called the **promoter**.

**Gene expression** refers to the timing and amount of protein production, which strongly influences plant function and development. Typically, the most important regulator of gene expression is the promoter. Genetic engineers typically alter the timing or amount of protein production by adding a new promoter to the gene that causes high expression.

The promoter and the structural gene may each originate from different genes and different organisms, and can be brought together in new combinations. For example, a promoter from a rice gene can be attached to a structural gene from a bacterium.

Some genes directly control the expression of several genes. The proteins produced by such genes are called **transcription factors**. Transcription factors sometimes have advantages for the engineering of genetically complex traits (such as NUE) that are controlled by several genes. But they can also affect the expression of genes that control traits other than the intended one—a result that may have undesirable consequences. Such a result can also occur if the expression of single genes that are not transcription factors is altered.

Altering gene expression has so far proved to be as important for improving NUE through GE as have structural genes. Most experimental increases in NUE have come from increasing the expression of existing structural genes (or similar genes from other organisms) rather than using genes that are fundamentally different from those already found in the crop.

## How We Evaluated GE's Prospects for Improving NUE

Ideally, to evaluate the efficacy of a new crop designed to increase NUE, we would study the plants as they are grown on a variety of working farms—in the field with varying soil conditions, plant densities, rainfall patterns (over a period of years), and other factors that influence plant growth. Such studies provide realistic estimates of commercial promise and reveal unintended consequences on and off the farm.

Because on-farm studies are costly, a series of preliminary, controlled, and more easily interpreted experiments are usually performed first. For example, new GE plants are typically evaluated first by growing them individually in pots in a greenhouse.

Laboratory and greenhouse studies have great value because they show how genetic manipulations manifest themselves in plants, rather than in a bacterium in a Petri dish. They do not, however, enable us to evaluate how a crop will contribute to a farming system that may retain or lose nutrients to the surrounding landscape, air, and water (see Box 2 for a discussion of different testing environments for GE plants).

The publicly available information on GE crops with NUE genes comes primarily from controlled studies conducted in growth chambers or greenhouses, and U.S. Department of Agriculture (USDA) records indicate that no such crops have yet been approved or commercialized. On-farm experiments, therefore, have not been conducted.

### Box 2. Methods Used to Study Crop NUE

The performance of new NUE crops may be assessed by growing them within structures or outdoors. The different methods have their own strengths and weaknesses: growth chambers provide the greatest control over growing conditions and the most precise comparisons, while commercial-scale studies provide the most realistic environment.

**Greenhouse and growth chamber studies** involve growing the experimental crop under highly controlled settings. Though greenhouses typically use ambient light and may not fully control temperature, they still represent an artificial environment compared with the exposed conditions of a crop field. Growth chambers are enclosed structures that typically control all aspects of crop growth including temperature, light, and humidity. Plants are often grown in pots rather than in groups or rows as on a farm.

**Field trials** test crops outdoors, but under conditions that can be monitored and treated in a controlled manner. Although field trials approach commercial crop production in terms of exposure to environmental conditions, they are much more limited in size (plots are often less than an acre), duration (often for only a few years), and geographic distribution.

**Commercial-scale studies** typically involve monitoring crop growth on commercial farm fields that are much larger than field trials, and may continue (continuously or intermittently) for many years. Commercial-scale studies may sometimes be performed like field trials, but at a much larger scale and for a longer duration.

Growth chambers and greenhouses cannot replicate the complex interactions between a plant and the environment that occur outdoors, including conditions that may lead to undesirable side effects. Field trials can begin to assess environmental effects, but sporadic phenomena such as pests and severe weather may not be present during the limited duration of a field trial.

Therefore, commercial-scale studies over a long period of time are needed to reliably detect the effects of sporadic, but important, environmental phenomena, as well as processes that take a long time to develop (such as the accumulation of organic nitrogen in the soil). Such studies may also provide considerable information about how plants affect each others' growth and about NUE, including nutrient loss from agricultural systems.

A relatively small number of field trials (which represent an intermediate step between growth chamber and on-farm studies) have been conducted, but the results of those trials—considered confidential business information—have not been released. Without comprehensive field studies, we cannot evaluate the promise of GE NUE crops under commercial conditions, or whether serious drawbacks such as impaired responses to drought or pathogens may emerge in the field.

Nonetheless, the available data provide a useful assessment of the state of development of GE NUE crops. Although many such crops appear to be in relatively early stages of development, and face several possible hurdles, there are a number of examples in the scientific literature (beginning in the 1990s, but primarily since 2000) of genes that have shown promise for improving NUE. Progress in this area mirrors our increased understanding of nitrogen metabolism by the genes involved in NUE, gained with the use of traditional genetic methods as well as tools from physiology and molecular biology (Hirel et al. 2007).

### Studies of GE NUE Crops

Researchers have focused much of their efforts to develop GE NUE crops on seven genes, primarily in major grain crops (rice, corn, and wheat) and the oilseed crop canola. Soybeans have been a common subject of USDA field trials for improved NUE, but the genes used in these trials are not known to the public. Most of the research in the public literature has centered on plant-derived genes important to nitrogen metabolism in plants, though some genes have come from bacteria (which resemble plants in some aspects of nitrogen metabolism). Many of these genes have been isolated and analyzed in experimental plants such as *Arabidopsis* as well as crops.

Genes that have been evaluated in the literature include:

- genes that code for nitrate and ammonium transporters that assimilate nitrogen from the soil;
- genes such as nitrate and nitrite reductases, which alter the form of nitrogen in the crop so it may be incorporated into organic (carbon-containing) molecules;
- genes that synthesize nitrogen compounds such as glutamine synthetase, which produces the amino acid glutamine (used to transport nitrogen through the plant); and
- genes responsible for remobilizing nitrogen from the vegetative parts of plants into the seed.<sup>1</sup>

The following discussion of studies described in the scientific literature focuses on those genes that have attracted the most attention and have shown the greatest promise for improving NUE.

In most cases, the GE strategy for nitrogen metabolism genes has been to boost their expression with gene promoters that cause the gene to be turned on at high levels in many plant tissues most of the time (Box 1) (Good, Shrawat, and Muench 2004). Boosting gene expression throughout a plant means that the protein product of gene expression will occur in plant tissues where it is not normally found, or in atypical amounts. This widespread change may increase the chance of undesirable side effects (or pleiotropy, discussed below).

Concern about the likelihood of unintended consequences stems in part from our understanding that most aspects of plant molecular biology (including nitrogen metabolism) are highly regulated and respond to changes in plant biochemistry. Therefore, atypical expression of nitrogen metabolism genes will likely cause some reactions by the plant. Whether these reactions will manifest themselves in plant growth and cause agricultural, environmental, or human safety problems is usually not entirely predictable given our current knowledge of plant biochemistry and metabolic networks (Sweetlove, Fell, and Fernie 2008).

<sup>1</sup> A more detailed list and discussion about these genes can be found in Good, Shrawat, and Muench (2004).

Using promoters that express nitrogen metabolism genes at high levels in many parts of the plant, in most cases, has resulted in increased NUE in experimental crops. Below and in Table 1 is a list of the gene-crop combinations of potential interest to genetic engineers.

Perhaps the most widely explored genes for improved NUE are those that control production of glutamine synthetase (GS). Several versions of these genes, called a “gene family,” appear to be central to nitrogen metabolism because glutamine is the primary compound involved in the movement of nitrogen throughout the plant, including into the growing seed. Versions of GS genes are found in the root and in the green parts of the plant. GS has been engineered into several crops.

*Glutamine synthetase in wheat.* GE wheat was developed using a bean GS gene and a strong promoter from a rice gene (Habash et al. 2001).

Plants were grown under controlled light and temperature in a growth chamber using a soil potting mix. The over-expression of this gene, compared with the normal wheat GS gene, in the green tissues of the plant resulted in an increased grain yield of about 10 percent, and increased grain nitrogen by a somewhat larger amount, under normal nitrogen fertilization. This occurs by increasing the reallocation of nitrogen in the plant from the leaves to the seed.

The root system of the GE GS wheat plants was also enhanced compared with non-GE wheat plants. While this may be a beneficial result, possibly enhancing nitrogen assimilation, it illustrates the side effects that often occur with the altered expression of engineered genes.

*Glutamine synthetase in maize.* A maize GS gene, normally expressed in leaves, was over-expressed using a promoter taken from a plant

**Table 1. Genes Used to Improve NUE through Genetic Engineering<sup>1</sup>**

Gene	Gene Source (Gene/promoter)	Engineered Plant	NUE Improvement <sup>2</sup> (Percent)	Grown in the Field? <sup>3</sup>
Glutamine synthetase (GS)	Bean/rice	Wheat	10	No
Glutamine synthetase (GS)	Corn/plant virus	Corn	30	No
Glutamate synthase (GOGAT)	Rice/rice	Rice	80	No
Asparagine synthetase (AS)	<i>Arabidopsis</i> /plant virus	<i>Arabidopsis</i>	21	No
Glutamate dehydrogenase	<i>E. coli</i> /plant virus	Tobacco	10	Yes
<i>Dof1</i>	Corn/plant virus	<i>Arabidopsis</i>	Nitrogen content: 30; growth: ~65	No
Alanine aminotransferase (ALA)	Barley/canola	Canola	40	Yes
Alanine aminotransferase (ALA)	Barley/rice	Rice	31–54	Yes <sup>4</sup>

Notes:

1 As reported in the public literature; other genes may be under private study by companies and universities.

2 Values for NUE are measured in different ways in different experiments. Therefore the values presented here are not directly comparable.

3 It is possible that field trials for these genes have been conducted but not disclosed to the public.

4 USDA field trials have been approved for this gene, but the results have not been reported to the public.

virus that produces GS in most plant tissues. Plants were grown in a greenhouse in pots, and produced about 30 percent more grain under low-level nitrogen fertilization (Martin et al. 2006).

**Glutamate synthase in rice.** Glutamate synthase (GOGAT) genes represent another gene family important in plant nitrogen metabolism, and have been used in experiments to improve NUE in rice. Genetically engineered indica rice—the primary subspecies grown in India and several other parts of Asia—was developed using an indica GOGAT gene under the control of a GOGAT promoter from a different rice subspecies, japonica rice (Yamaya et al. 2002).<sup>2</sup> Grain yields for GE indica plants grown in pots in controlled conditions were 80 percent higher than for the non-GE indica plants.

**Asparagine synthase in *Arabidopsis*.** As with the GS gene, the asparagine synthase (AS) gene controls the synthesis of an amino acid that can be important for transporting nitrogen through a plant. AS was over-expressed in the experimental plant, *Arabidopsis*, using a strong promoter from a plant virus that produces high levels of AS in most plant tissues (Lam et al. 2003). The GE plants were grown in pots under controlled light and temperature and normal levels of nitrogen. Seed protein content increased by about 21 percent.

**Glutamate dehydrogenase in tobacco.** Under field conditions in Illinois, a bacterial glutamate dehydrogenase gene (from *E. coli*) expressed at high levels in tobacco using a promoter from a plant virus produced up to about 10 percent more plant biomass than the non-GE plants over a period of three years (Ameziane, Bernhard, and Lightfoot 2000). Increased crop yield appeared to occur only at normal nitrogen fertilization levels.

**Dof1 transcription factor in *Arabidopsis*.** The maize *Dof1* gene is a transcription factor (Box 1) that controls the expression of several genes involved in carbon metabolism (Yanagisawa et al. 2004). Carbon and nitrogen metabolism are linked in plants, and because many plant molecules

contain significant amounts of both carbon and nitrogen, increased expression of a gene for carbon compounds may also boost nitrogen in the plant. The GE *Arabidopsis* plants containing *Dof1* at high levels accumulated more nitrogen than normal plants—in some cases more than twice as much—when grown in the laboratory on an artificial agar-based medium containing low amounts of nitrogen. The GE plants also showed greater growth than their non-GE counterparts, although the amount of growth difference was not quantified.

**Alanine aminotransferase in canola.** The alanine aminotransferase (ALA) gene is one of the few nitrogen metabolism genes that has been expressed from a promoter restricted to specific plant tissues and environmental conditions, and grown in the field rather than only in greenhouses or growth chambers. Investigators combined a barley ALA gene with a promoter that functions in the roots of canola plants and used the resulting combination to genetically alter canola plants (Good et al. 2007).

In field trials over a two-year period, and with nitrogen fertilizer application rates 40 percent below normal, they observed canola seed yields equivalent to those achieved at typical soil nitrogen levels. At more typical application rates, the GE canola exhibited a yield increase of approximately 33 percent. At high application rates (280 kg/hectare), no yield advantage was reported.

**Alanine aminotransferase in rice.** A barley ALA gene was expressed by a root-tissue-specific promoter in GE rice (Shrawat et al. 2008). Under controlled conditions, grain yield increased between 31 and 54 percent compared with the non-GE rice. Root and fine root biomass also increased considerably, as did nitrogen uptake. The USDA has also approved field trials of ALA rice, but the results have not been released to the public.

**Summary.** Our review of the literature revealed several genes important to plant nitrogen metabolism that have drawn the interest of genetic engineers. Of these, GS genes have probably attracted

<sup>2</sup> There are several distinct types of Asian rice—indica, japonica, and javanica—all of the species *Oryza sativa*, and all generally inter-fertile. Indica rice varieties are the most widely grown.

the widest interest. Promising results have also been observed with GOGAT and ALA. Work on the latter appears to be the most advanced, with field trials lasting several years (see below).

The studies described above, mostly conducted in controlled environments, demonstrate that NUE genes can increase both seed yield (at low, normal, or high nitrogen fertilizer levels) and plant nitrogen content. Grain yield increases in greenhouse tests have ranged from approximately 10 percent to 80 percent (Table 1). However, tests in controlled environments may not identify undesirable genetic side effects that manifest themselves under certain environmental conditions, and may not detect other limitations imposed by commercial-scale crop production.

### Approved Field Trials of GE NUE Crops

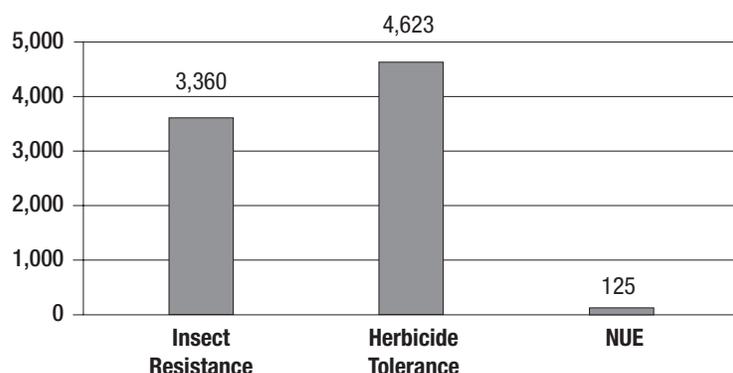
Field trials test experimental GE crops under conditions that may approach those on farms, and afford the opportunity to assess a variety of possible environmental impacts as well as NUE at the scale of a crop field (rather than an individual plant). However, secrecy about genes and field trial results greatly limits our ability to evaluate the prospects of these genes. Field data are critical to assess the success of efforts to produce high-NUE

crops because, for example, an individual plant may have high NUE when grown in a pot but lower NUE in the field if fertilizer is applied before root systems have developed sufficiently to colonize most of the field's soil. Nutrient losses often depend on the timing of not only fertilizer application but also irrigation and/or rainfall.

U.S. field trials of GE crops must receive USDA approval, and are listed in the USDA's publicly available GE field trial database. This database therefore provides the number of all approved NUE field trials in this country, and offers a general sense of how advanced this research is compared with other GE traits.

Between 1987, when the USDA initiated its field trial program, and 2000, only 26 field trials for nitrogen metabolism were approved, but 99 have been approved since then (Animal Plant Health Inspection Service 2009). This substantial increase over the past decade suggests growing interest in, and identification of, possible NUE genes. Nevertheless, the total number represents only a fraction of the field trials approved for insect-resistant and herbicide-tolerant GE crops: there have been 4,623 field trials approved for herbicide tolerance and 3,360 for insect resistance through 2008 (Gurian-Sherman 2009) (Figure 3).

**Figure 3. USDA-Approved Field Trials of GE Crops\***



\* Field trials for herbicide tolerance and insect resistance approved through February 2009. Field trials for NUE approved through August 2009. Source: USDA, APHIS Biotechnology Regulatory Services, online at [www.isb.vt.edu/cfdocs/fieldtests1.cfm](http://www.isb.vt.edu/cfdocs/fieldtests1.cfm).

The relatively small number of field trials for NUE shows that advances in this research are more recent than that on other traits, and less advanced. It is also consistent with the small number of genes the public literature suggests have attracted the most interest. For example, if the number of field trials for the ALA gene is representative of other NUE genes, then dividing the total number of NUE trials (125) by ALA field trials (17) suggests about seven NUE genes being studied in field trials. On the other hand, it is also possible that some trials may involve several NUE genes.

All of the field trials conducted through 2004—as well as several conducted afterward—use the general term “nitrogen metabolism altered” to describe the GE trait. It is unclear how many of these 60 approved field trials were attempting to increase NUE specifically, but because the terms “nitrogen metabolism altered” and NUE are used to describe the same gene at different times, we have included these trials in our total under the assumption that at least some had the goal of improving NUE.

The USDA database also provides a window on which institutions are investing in enhanced NUE via GE, and which crops have received attention. The large majority of field trials, for example, have been conducted by either Monsanto or Pioneer Hybrid. Several have also been conducted by Arcadia, which is using the ALA gene. This company appears to be collaborating with Monsanto, as revealed by a paper discussing GE ALA in canola that was co-authored by scientists employed by both companies (Good et al. 2007). Most of the NUE field trials involve corn, with many involving soybeans, canola, and rice as well. A few have been conducted using other crops, but have not been carried forward to recent years; one involving the potential biofuel crop switchgrass was approved in 2009.

Because of current limits on the public availability of field trial data, we must rely on inferences about the genetic and physiological effects

of GE NUE genes on the plant to evaluate their prospects for success.

### **Possible Risks Related to GE NUE Genes**

Limited testing has already revealed several possible undesirable or harmful unintended changes in the expression of plant genes due to the engineering of NUE genes. Even when GE NUE crops show promise in greenhouse tests, the possibility of undesirable or harmful side effects (pleiotropic effects) when those crops are grown in the field may reduce the value of the gene. Field trials conducted for several years are more likely to detect undesirable side effects, but some may only be observed in response to occasional occurrences, such as extreme heat or cold or an outbreak of pathogens, that may not occur during field trials.

One particularly worrisome side effect of GE NUE genes is that they may indirectly increase the production of harmful substances in the edible parts of crops. Most crops have genes that produce harmful substances, but these genes are not expressed, or are expressed at low levels, in the edible parts of crops. Engineered genes, however (or genes manipulated through traditional breeding), may have the opposite effect due to complex interactions between the engineered gene and crop genes (National Research Council 2000).

Consider the *E. coli* glutamate dehydrogenase gene, which was studied as a possible NUE gene (Ameziane, Bernhard, and Lightfoot 2000). When expressed in tobacco it altered the production of many plant compounds (some were increased and some were decreased), most notably the amounts of nine known carcinogens and 14 potential drugs (Munger et al. 2005). Although tobacco is not edible, this example illustrates the possibility of unpredictable and potentially harmful changes in food crops.

Because we know that the nitrogen status of plants affects various aspects of their physiology, including defense against pests (Craine et al. 2003; Vitousek et al. 2002), it is reasonable to ask

whether altering nitrogen metabolism with NUE transgenes could influence the amounts and types of important plant components.

Recent tests have found that overexpression of ALA in rice causes a significant change in the expression of 91 other genes in the roots and shoots of rice plants grown hydroponically (Beatty et al. 2009). Seventeen of these genes had altered expression in two independently created ALA rice plants. The identified rice genes are involved in various aspects of plant function: several have been associated with defense against pathogens, one of which (called the osmotin-like, thaumatin-like gene) was expressed at a two- to three-fold lower level than in normal rice plants. Two genes of the PR10 type (a “pathogenesis-related” protein implicated in the defense of plants against disease) were also found to have significantly reduced expression. Reduced expression of these genes raises a question about the possible increased susceptibility of ALA GE rice to disease.

In summary, pleiotropic effects are a distinct possibility for GE NUE crops, but have yet to be explored in the public literature. Because they are largely unpredictable and may only occur under specific environmental conditions, these side effects may not be revealed by the types of experiments thus far performed (mostly under greenhouse conditions). Even when such crops are grown in the field, some changes in gene expression may only be detected through sophisticated testing of plant genes or compounds, as was done for tobacco containing a glutamate dehydrogenase gene and rice containing an overexpressed ALA gene; such testing is not required under current U.S. regulations. Many side effects may be harmless or inconsequential for crop production, but the possibility that some could be undesirable should be carefully evaluated.

### **Commercialization of GE NUE Crops**

There is not enough detailed information about the performance of GE NUE crops at this time to

clearly understand their prospects for commercialization. Commercial potential is therefore generally inferred from available information about a) the efficacy of NUE genes and b) possible hurdles that may be faced as these crops are tested under more realistic conditions and as they proceed through the regulatory process.

The NUE values obtained for GE crops in recent tests, most of which were conducted in controlled environments and with limited durations, are unlikely to be maintained on commercial farms under real-world conditions. In addition, the apparently limited number of comparisons with existing crop varieties that may differ in NUE also suggests that NUE values for GE crops may be lower than reported (see Chapter 3).

Only the actual performance of GE NUE crops will determine whether these varieties are economically viable and attractive compared with other technologies for improving NUE. The NUE values of GE crops need to be high enough to justify the costs of development, production, and marketing, as well as the extra costs farmers must pay for GE seed.

Undesirable side effects, where they exist, may reduce the efficacy of these crops, force farmers to pay additional costs, and affect how widely the crops are adopted if approved. For example, if plant diseases are exacerbated in some instances (see above), higher costs for disease control could reduce the adoption rate of the crop and, in turn, the practical impact on NUE. When side effects are harmful to the environment, they may also prevent regulatory approval.

The ALA gene shows the most promise for commercialization based on publicly available information. It is the only gene identified in USDA field trials, 17 of which—almost 14 percent of all NUE field trials—have been approved since 2002. This long record suggests that the ALA gene may be approaching the late stages of testing. On the other hand, the lack of large-scale field trials—none of more than five acres—that are

usually conducted within several years of regulatory approval may suggest that commercialization is at least several years away.

This gene also reduced expression of several genes in rice that help the plant defend itself against disease. It is unclear at this time what practical effects this may have.

No petition for deregulation of any NUE crop—a prerequisite for commercialization—has yet been announced by the USDA in its public database (Animal and Plant Health Inspection Service 2009b). Examination of the petition database shows that deregulation decisions generally require at least two years. It seems unlikely

therefore that any NUE crop will be commercialized in the United States before 2012.

Most of the GE NUE crops reported in the scientific literature appear to be at relatively early stages of development, with the possible exception of the barley ALA gene in canola and rice. If other genes are in more advanced stages of development, the work is occurring behind closed doors. Based on the information available to us, the prospects for commercialization of GE NUE crops must be considered largely uncertain at this time. These prospects should be compared with those of other methods and technologies for addressing nitrogen pollution, which are addressed in the next chapters.

## CHAPTER 3

# *Improving NUE through Traditional and Enhanced Breeding Methods*

---

**T**raditional breeding involves the controlled mating of plant parents selected for their desirable traits. This effective technology, responsible for most genetic improvement in crops over the last 100 years, can now be enhanced by new genomic technologies that assist scientists in identifying prospective genes and parents. GE is sometimes considered a form of breeding, but it is distinct from previous crop breeding methods and is not referred to as breeding here. Genomic breeding methods use knowledge gained from the study of plant genes, but is not a form of GE and uses plant-mating methods similar to traditional types of breeding.

Information on traditionally bred crops is difficult to obtain because there is no registry of such crops. Some information can be found in the scientific literature and by talking to plant breeders at public institutions.

Traditional breeding has a strong track record in conferring important new traits on crops (e.g., disease and insect resistance, drought tolerance, dramatically increased yields), and there is every reason to think it would be able to achieve success in improving NUE. Like GE, traditional breeding uses genetic variation as a means to improve crops, but unlike GE, which can derive that variation from unrelated organisms, traditional or genome-enhanced breeding methods exploit variation within the crop species or its sexually compatible wild relatives.

### **NUE Improvements in Commercial Varieties**

Increases in NUE through traditional breeding for wheat and oats have been demonstrated over

the second half of the twentieth century in Europe (Muurinen et al. 2006; Brancourt-Hulmel et al. 2003). For example, Brancourt-Hulmel et al. grew wheat cultivars introduced between 1946 and 1992 under the same conditions, including amount of added nitrogen. Comparing the average yield at high nitrogen input for the four varieties introduced between 1946 and 1964 with the four introduced between 1987 and 1992, the study data show a 29 percent gain in NUE over the approximately 35 years between the average introduction dates of the two periods (1955 and 1990).

Also, a study comparing older and newer varieties of wheat in Mexico between 1950 and 1985 (with a typical amount of applied nitrogen) showed a yield increase for the newer varieties of 60 kilograms per hectare per year (Ortiz-Monesterio et al. 1997), or about 42 percent in 35 years. Although these studies represent just a small sample of the grain varieties developed in recent decades, the findings suggest that yield gains in other grains over the past 50 years may also contribute to improvements in NUE.

### **The Impact of Higher Yield on NUE**

Success in improving crop yields often improves NUE as well. In the United States, nitrogen use has been roughly constant from the 1990s through the mid-2000s (Wiebe and Gollehon 2006b), while the yield of major grains has increased: about 13 percent for wheat, 16 percent for soybeans, and 28 percent for corn over the past 13 years (Gurian-Sherman 2009). Historically, about half of U.S. crop yield gains have been attributed to

crop breeding (Duvick 2005). If that relationship has continued during the past dozen years, then traditional breeding may account for roughly half of the improvement in NUE over this period based on yield per unit of added nitrogen, or about half of the increased yield value.

This provides a very rough estimate of traditional breeding's contribution to improved NUE in this country in recent years. Overall, considerable improvement has occurred over the past several decades, as a result of both breeding and other means (Table 2).

### Genetic Variability of NUE-Related Traits in Major Crops

As past studies suggest, there is considerable potential for improving NUE through traditional breeding methods, but this potential depends on the variability of NUE-related traits, and their corresponding genes, within a crop or its wild relatives. Much of the genetic potential of major crops remains untapped for many traits (Hoisington et al. 1999), which likely include NUE.

Hoisington et al. found that only a small portion of the genetic variation in corn and wheat has been utilized in current crop varieties. This under-utilization is especially true for wild relatives such as *Tripsacum* and *Teosinte* species for maize improvement and *Aegilops*, *Agropyron*, and non-wheat *Triticum* species for wheat improvement. Only about 1 percent of the U.S. maize germplasm base, and only about 5 percent of the globally available germplasm base, has utilized these resources (Hoisington et al. 1999). For wheat, only an estimated 10 percent of varieties as of 1986 may have used the genetic resources from exotic wheat varieties (called landraces) to improve existing wheat varieties.

Despite this minimal use of the available genetic diversity, tremendous contributions have already been made to maize and wheat improvement, including numerous genes for disease resistance, insect resistance, stress tolerance (such as for drought), quality traits, and yield (Hoisington et al. 1999)—suggesting there is also potential here for improving NUE. Another possible resource is

**Table 2. Improvements in Nitrogen Use Efficiency**

Crop	Time Frame* (Years)	Country	Source of NUE Gain	NUE Gain (Percent)	Reference
Wheat	35	Mexico	Breeding	42 (59 kg/ha/year)	Ortiz-Monesterio et al. 1997
Wheat	35	France	Breeding	29	Brancourt-Hulmel et al. 2003
Rice	~15	Japan	Unknown	32	Dobermann and Cassman 2005
Maize	~20	United States	Unknown	36	Dobermann and Cassman 2005
Cereal crops	15–20	United Kingdom	Unknown	23	Dobermann and Cassman 2005

\* All studies were conducted in the second half of the twentieth century.

the existence of sexually compatible wild relatives for virtually all other major food crops, such as the rice relative *Oryza rufipogon* and the soybean relative *Glycines soja* (Ellstrand 2003). NUE has not been widely investigated in these genetic resources.

Some research with nitrogen metabolism genes shows some of the genetic variability for traits or genes associated with NUE within a crop species. Perhaps most striking is the genetic variability found in rice for a GOGAT gene (Yamaya et al. 2002). The gene used for this experiment (or more properly, the gene promoter) originated in one type of rice (japonica) and was inserted into another type of sexually compatible rice (indica) after being attached to the indica GOGAT gene, resulting in an 80 percent increase in yield. Because the genes were from sexually compatible varieties of rice, a similar yield improvement may be accomplished using traditional breeding techniques.<sup>3</sup>

Related research shows a similarly high level of variation in the amount of another gene and protein widely studied and used to develop GE NUE crops. GS protein in rice leaves ranged from 2.55 to 16.18 micrograms—more than a six-fold difference—in the offspring of two varieties, one a japonica and the other an indica type (Obara et al. 2001). To the extent that levels of GS expression are important for improving NUE, as has been seen in other experiments, this variation suggests substantial potential for improving NUE through breeding.

Obara et al. identified this variability by comparing only two varieties of rice, albeit varieties that are genetically divergent. These two varieties do not contain all of the genetic variability contained in all rice varieties or their wild relatives, and therefore do not reveal how much additional variability—which may be used to increase NUE—could be found if more of the rice gene pool was examined.

Research in corn has revealed numerous chromosomal locations associated with variation in different aspects of NUE (Coque and Galais 2006; Galais and Hirel 2004). Several of these regions, called quantitative trait loci (QTL), are also associated with genes that have been used experimentally in GE to enhance NUE, including several GS genes and glutamate dehydrogenase. Genetic markers that are linked to QTL and function as genetic fingerprints can be used to track the QTL during breeding in a process called marker-assisted breeding, which can greatly accelerate breeding.

QTL and significant genetic variability for NUE have also been found in barley (Mickelson et al. 2003). Preliminary work in wheat has identified several QTL associated with NUE, which include one or more GS genes in the flag leaf (the leaf nearest the wheat seed head), which is known to be important for grain yield (Habash et al. 2007). Development of improved crop varieties that use QTL can be difficult to accomplish in practical breeding programs because they may perform well in one environment or in one variety of the crop, but not as well in others (Bernardo 2008; Dekkers and Hospital 2002; but see Heffner, Sorrels, and Jannick 2009 for a more optimistic view).

Higher values for NUE-associated traits have been observed in progeny than in either parent (Mickelson et al. 2003).<sup>4</sup> This demonstrates that, at least for the varieties tested, improvement of NUE-associated traits found in parent varieties is possible. Traditional breeding's ability to improve NUE can thus be enhanced by new methods based on the identification of specific genes or regions of crop genomes.

Even if there is considerable genetic diversity within a crop, however, it is possible that current commercial varieties may already contain very good genes for NUE, which could reduce the

<sup>3</sup> Traditional breeding would not provide exactly the same result, because the promoter from the japonica rice gene was combined with a structural gene from indica rice using GE, which would not typically occur with traditional breeding. We are assuming that overexpression (rather than some other activity) of the GOGAT enzyme is primarily responsible for the results, but it is possible that overexpression is not responsible for the entire difference in NUE.

<sup>4</sup> This is a characteristic called transgressive hybridization, which may be associated with strongly adaptive traits. Many traits in the progeny of varietal crosses, however, show values between those of the two parents.

potential for further improvement through traditional breeding. Other possible barriers include undesirable pleiotropic effects similar to those that may occur through GE.

The potential for improving NUE through traditional breeding is likely to be considerable (but not unlimited). Recent research on crop genetic variability, and variability for NUE traits specifically, suggests considerable variation exists for traits associated with NUE. The extended crop gene pool that includes sexually compatible wild relatives does not seem to have been explored for the purpose of improving NUE, but may provide additional opportunity to improve NUE through breeding.

### **Strengths and Limitations of Breeding Compared with GE**

Given the need to allocate public research money judiciously, it would be wise to compare the relative prospects of GE and traditional breeding for improving NUE. In general, both methods have the capacity to generate improved crop varieties, but only traditional breeding has thus far succeeded in bringing varieties with improved NUE to the marketplace.

For both GE and breeding, reported values of improvement in NUE should be viewed as preliminary prior to extensive field testing that includes comparisons with the best current varieties of the crop. Many of these values were derived from studies of plants grown in pots, inside growth chambers or greenhouses where light, water, and temperature were controlled. Growing conditions in the field can be expected to introduce stresses and other environmental effects that may negatively affect NUE values. In addition, the genetic and physiological complexity of nitrogen metabolism in plants presents a considerable challenge for GE approaches relying on single genes (Lawlor 2002), which may lead to problems in the field.

Equally important, the values reported in the literature for GE NUE crops are typically determined by comparing the GE variety only with

its non-GE progenitor. Such comparisons do not reflect the variation in NUE that exists in commercial or other available varieties of the crop. Thus, because some other varieties of the crop may deliver better NUE than the one used for comparison with the GE variety, the relative NUE advantage of the engineered gene would be less than the value reported in the literature.

In theory, GE should be capable of developing new crop varieties more quickly than breeding (Long et al. 2006) because it involves adding only one or a few genes, while breeding combines the entire genomes of the two parents. Removing undesirable genes to arrive at the desired combination of genes typically requires years of breeding.

But, this presumed advantage of GE appears to be minimal or absent in practice. First, the GE process itself introduces mutations and other changes in the plant that may be undesirable (National Research Council 2004). Although plants are initially screened for obvious unintended alterations, many potential changes can involve plant metabolism or occur only under certain environmental conditions—factors that would not be detected during the initial screening. Many of these mutations can be eliminated by the same kind of iterative process used to improve plants through traditional breeding, but this requires considerable time.

Time is also added to the GE process because the effects of new engineered genes under different growing conditions are not predictable. New GE varieties must therefore be grown in field trials for several years (as must new varieties developed through traditional breeding).

Meanwhile, the breeding process has been improved by our increasing understanding of plant genetics, physiology, and biochemistry. This has led to selection methods that can accelerate the breeding process substantially, further reducing any advantages of GE.

Certain studies have found that GE crops require more than a decade to be developed and

deployed—similar to the amount of time needed for traditionally bred crops (Goodman 2004; Gepts 2002).

Finally, as noted above, the supposed advantage of GE over breeding in providing expanded access to genetic resources has yet to result in improved NUE. The available research papers that have provided preliminary quantification of NUE improvement through GE or breeding have not revealed a distinct advantage for either approach.

To reiterate, although GE and traditional breeding both have the potential to produce new crop varieties with higher NUE, only traditional breeding has succeeded in bringing such varieties to market (GE's attempts are limited to the past

10 to 12 years). Whatever advantages GE is presumed to have in generating new varieties, they are not apparent in this arena or any other involving complex traits.

So, while there is no reason to abandon ongoing GE efforts, there is no reason to expect more from them than traditional breeding, and they should not be favored in the allocation of scarce resources. Evidence shows that public resources for traditional breeding have declined globally in recent decades (Kloppenburg 2005) despite its success with complex traits such as NUE. We must ensure that public sector traditional breeding receives a level of support commensurate with its demonstrated potential.

## CHAPTER 4

### *The Ecosystem Approach to NUE*

---

**E**cosystem approaches consider the spatial, temporal, and species interactions that can affect a crop's NUE—factors not necessarily considered during breeding for NUE, which often focuses single-mindedly on crop yield per unit of added nitrogen. Viewed exclusively through this crop production lens, NUE may miss important routes of nitrogen loss from the farm. For example, a crop with improved NUE may not reduce nitrogen losses early in the growing season, prior to vigorous crop growth and root production. Ecosystem approaches thus represent a possible route to both higher crop yields and lower nitrogen loss and pollution.

#### **A Big-Picture Perspective**

Ecosystem scientists view cover crops as part of a holistic plant-soil system, and their approach to measurement reflects this view. Key data points often include actual losses of reactive nitrogen from the farm, in the form of runoff, leaching into groundwater, and gaseous emissions from the soil (e.g., Tonitto, David, and Drinkwater 2006; Drinkwater, Waggoner, and Serrantonio 1998), and sometimes include a crop's uptake of nitrogen as a percentage of the nitrogen applied. Through this lens, NUE could be defined as the amount of plant matter or grain produced with the least nitrogen pollution.

An ecosystem perspective also expands the time scale over which we consider NUE, drawing attention to periods when plants are not actively growing or when recently planted crops have immature

root systems that draw nitrogen from only a small portion of the total soil volume. Reactive nitrogen that goes unused when crops are not actively growing can be a major source of nitrogen loss from farms (Tonitto, David, and Drinkwater 2006). Therefore, crop species that can be planted earlier in the season—or that persist later in the growing season—can potentially reduce nitrogen loss by capturing more soil nitrogen than crops with a shorter growing season. This intersection between root development, a plant's nitrogen demand, and the timing of fertilizer application also plays a role in determining farm-level NUE.

Viewing the agricultural system at large spatial and temporal scales points to a variety of approaches (precision agriculture, use of cover crops) that can control the flow of nitrogen between farm and adjacent systems (air, water). Cover crops, which are often used in organic or similar agricultural systems, and precision farming, which is used more often in traditional systems, are discussed in Chapter 5.<sup>5</sup>

#### **The Time Is Ripe for a New Approach**

Any progress in nitrogen use we have made up to this point has not led to the decrease in nitrogen pollution we need. For example, U.S. corn yields have increased about 28 percent over the past 13 years (Gurian-Sherman 2009), and productivity of other major crops such as soybeans and wheat have also increased. Nitrogen fertilizer use on major crops remained about the same during most of that period (Wiebe and Gollehon 2006a),

---

<sup>5</sup> Means of reducing nitrogen pollution directly (e.g., planting vegetative buffer strips between crop fields and streams) are also important but not covered in this report.

suggesting a substantial improvement in NUE—but several indicators suggest nitrogen pollution has not improved significantly.

For example, the so-called dead zone in the Gulf of Mexico, largely the result of agricultural nitrogen pollution, expanded during the 1990s, peaked in 2002, and has remained at near-record size since. The U.S. Environmental Protection Agency (2008) has suggested that nitrogen pollution will need to be reduced about 45 percent to substantially shrink the dead zone. Other studies confirm that nitrogen

pollution remains a serious problem (Rockström et al. 2009; Vitousek et al. 2009).

Looking to the future, this analysis suggests that simply increasing the efficiency of crops (as defined by yield per unit of nitrogen applied) is unlikely by itself to reduce pollution sufficiently. Any improvements in NUE must therefore be viewed from an ecosystem perspective that gives equal weight to preventing nitrogen loss and increasing crop yields.

## CHAPTER 5

*Other Means of Improving NUE*

In addition to GE and traditional breeding, several other agricultural technologies or practices show promise for improving NUE. This chapter sets our evaluation of GE and breeding in a broader context by providing a brief overview of prominent alternatives for improving NUE: precision farming and organic or other “low-external-input” farming systems<sup>6</sup> that use livestock manure, or “green manure”<sup>7</sup> from cover crops,<sup>8</sup> as sources of crop nutrients. Both precision farming and cover crops can be incorporated into industrial agricultural systems; systems that use little or no pesticides or synthetic fertilizers require a more fundamental change from the predominant industrial farming system, but deliver a richer set of environmental benefits.

Both precision farming and organic or similar systems attempt to improve NUE by managing nitrogen input and the amount of nitrogen in the soil rather than altering the plant genome. Precision farming focuses on matching the nitrogen supplied from synthetic fertilizers to the needs of the crop, avoiding the excesses that contribute to nitrogen pollution. Organic farming and similar systems emphasize building soil quality and soil organic matter, which provides multiple benefits including reduced nitrogen loss from the farm.

In general, the negative environmental impacts of nitrogen, including air and water pollution and the production of nitrous oxide, increase as the amount of inorganic nitrogen applied increases.

Industrial agriculture, which commonly applies a large amount of synthetic, inorganic reactive nitrogen at once—more than crop roots can assimilate over a short period of time—is especially damaging. By contrast, methods that minimize the use of synthetic fertilizer, release nitrogen slowly over the growing season, or remove excess nitrogen from the soil reduce the negative impacts of nitrogen.

Both organic and precision farming take into account the nitrogen sources already available in soil (so-called indigenous nitrogen), which is primarily organic (i.e., bound to carbon atoms) in form. Organic nitrogen breaks down into inorganic forms that are used by the crop but can cause pollution if they find their way into water or air.<sup>9</sup>

It is generally desirable to increase the amount of indigenous nitrogen available as a source of inorganic nitrogen for crop nutrition because it tends to contribute less to nitrogen pollution (Cassman, Dobermann, and Walters 2002). Indigenous nitrogen generally releases inorganic nitrogen continuously, in amounts smaller than industrial agriculture’s typically large applications of synthetic fertilizer.

The amount of organic nitrogen in the soil and the rate at which inorganic nitrogen is applied to the soil or released from organic sources are important considerations for both organic and precision farming. Specifically, the amount of synthetic inorganic nitrogen added to the soil should take into account the amount released from the indigenous

6 Low-external-input systems emphasize the use of biological principles to achieve soil fertility and pest control, and include organic farming as well as methods that allow a minimal use of synthetic fertilizers or pesticides.

7 Green manure refers to the use of plants as a means of supplying nutrients to other crops. Green manure crops are often grown during seasons when food crops are not grown; instead of being harvested they are plowed into the soil, where they release their nutrients.

8 Cover crops are planted to protect soil that would otherwise lay bare (between cropping seasons, for example) and subject to erosion. Cover crops also take up inorganic nitrogen that would otherwise be lost from the field. Plowing cover crops into the soil prior to the planting of cash crops provides nutrients, improves soil quality, and increases soil carbon content.

9 Some organic compounds can be used by crops but are not as important as inorganic forms. Some can also move through soil into groundwater, but these are also generally unimportant.

nitrogen supply. Because this can be challenging in practice, it is not always done.

## Cover Crops

Nitrogen can be supplied to crops by incorporating livestock manure or leguminous plants used as green manure into the soil. Both kinds of manure contain organic forms of nitrogen incorporated into large molecules such as proteins that are broken down into the smaller inorganic forms useful to crops.

Many of the major crops that are the target of both GE and traditional breeding cannot produce useable nitrogen, but others—legumes, for example—can. Legumes include important food and feed crops such as beans, peas, soybeans, peanuts, and alfalfa, as well as cover crops such as vetches and clovers. These crops live in close association with bacteria that can produce reactive nitrogen usable by the crop itself<sup>10</sup> and by non-legume crops planted in succeeding seasons. Because legume cover crops may supply most or all of the nitrogen needed for subsequent crops to produce high yields, incorporation of legumes into agricultural systems can reduce the need to supply synthetic nitrogen (thereby helping to reduce nitrogen pollution).

Legumes supply nitrogen in the form of organic molecules that are generally retained in the soil for longer periods of time than synthetic nitrogen—an additional advantage for reducing pollution. But because much of the organic nitrogen may be converted into more reactive forms such as ammonia or nitrate relatively quickly under certain conditions, the organic sources must be properly managed to avoid causing nitrogen pollution.

Manure and green manure also add carbon and other nutrients to soil, which may generally improve soil quality. For example, increasing soil organic matter generally improves the soil's water-holding properties and soil nitrogen levels, thereby improving the ability of crops to survive drought

(Lotter, Seidel, and Liebhardt 2003). Use of cover crops on otherwise fallow soil also greatly reduces erosion and may remove heat-trapping carbon dioxide from the atmosphere (Teasdale, Coffman, and Magnum 2007; Pimentel et al. 2005; Drinkwater, Waggoner, and Serrantonio 1998). Public policies aimed at improving NUE should therefore consider both the positive and negative impacts of the practices involved.

A long-term study in Pennsylvania comparing industrial agriculture practices with those of organic farming found that the latter produced much less leaching of nitrate into groundwater. The organic system did not use insecticides, herbicides, or synthetic fertilizers and relied on either legume cover crops—grown from fall to spring when the cash crops, corn and soybeans, were not growing—or manure to supply organic nitrogen (Drinkwater et al. 1998). Despite the fact that the organic and industrial systems used similar amounts of added nitrogen, considerably more nitrogen was retained in the soil of the organic system, which also lost about 50 percent less nitrogen to leaching through the soil (a potential source of water pollution). Yields for the organic crops were about 9 percent lower than the industrial crops, so the net nitrogen savings were considerably higher in the organic crops on a per-unit basis.

Soils from long-term organic farming systems have shown higher overall soil organic matter and organic nitrogen levels than industrial agriculture systems (Mariott and Wander 2006). Furthermore, organic systems have produced 40 percent more particulate matter (organic matter in an intermediate stage between fresh plant matter and decayed matter), which is associated with the ability to slowly release nitrogen that may be used by crops.

A meta-analysis<sup>11</sup> of 35 research projects examining nitrogen leaching and yield found dramatic reductions in leaching from fields incorporating cover crops compared with fields that did not (Tonitto, David, and Drinkwater 2006). This

<sup>10</sup> Legumes have a symbiotic relationship with particular types of bacteria that live in root structures called nodules and convert nitrogen into forms the crop can use for nutrition.

<sup>11</sup> A meta-analysis determines the combined statistical significance of many separately conducted research projects.

occurred despite the fact that many of the research projects, used for comparison with fields incorporating cover crops, included conservation tillage—associated with improved soil properties—as part of their industrial agricultural practices.

In rotations with non-legume cover crops, the cash crop was fertilized with synthetic fertilizer; in rotations with legume cover crops, the legume provided the nitrogen for subsequent cash crops. The cover crops were typically planted in the fall and plowed into the soil in the spring, prior to planting the cash crops. Non-legume cover crops such as rye reduced nitrogen leaching by an average of 70 percent compared with industrial crops, without reducing cash crop yields, while legume cover crops reduced leaching by 40 percent and averaged 7 percent lower cash crop yields (about 10 percent lower for grain crops).

Yields from the cover-cropped systems tended to be lowest in more northerly areas where the cover crops tended to produce less plant material and thus less nitrogen. In important agricultural areas where the cover crops tended to grow well—and thereby produced amounts of nitrogen comparable to synthetic nitrogen used to grow the industrial cash crops—yields were essentially the same.

Similar yield results were also found in another recent meta-analysis (Badgley et al. 2007). These results challenge the assertion that nitrogen from legumes is much less capable of producing the yields that can be achieved with synthetic nitrogen fertilizers (Smil 2000).

Cover crop systems do have several limitations that could benefit from greater research (Snapp et al. 2005). For example, cover crop growth, and hence their contribution to NUE or nitrogen fertilization, depends on the weather; low rainfall or cold autumn temperatures can reduce the growth of common crops, and nitrogen production of legume cover crops. Winter rye grown as a cover crop in Minnesota was effective in reducing nitrogen leaching in only one of four years studied

(Strock, Porter, and Russelle 2004), but still reduced nitrate loss by an average of 13 percent.

The seed, planting, and incorporation of cover crops into the soil involve expenses and farming challenges that must also be taken into account. Incorporating the cover crop at the appropriate interval before cash crop planting, for instance, can sometimes be a problem. Under certain conditions, such as heavy rainfall after legume cover crop incorporation but prior to vigorous cash crop growth, the cover crop may contribute to nitrogen loss.

### **Precision Farming**

The pattern, timing, and amount of fertilizer applications makes a significant difference in how much pollution will be caused by reactive nitrogen. Synthetic nitrogen fertilizer is often applied once at the beginning of the crop growing season or the preceding autumn, in an amount too large to be entirely assimilated by crop roots before some is lost. The basic premise of precision farming is to apply fertilizer in amounts sufficient to attain the desired yield without exceeding the amount the crop can utilize.

One practice being used by many farmers to more closely match nitrogen supply to crop need is to split fertilizer applications between the beginning of the growing season and later in the year. Another practice that improves NUE is fertilizing in the spring instead of the fall. Fall nitrogen application, especially when cover crops are not planted, allows considerable nitrogen loss to occur prior to crop growth in the spring. These methods have probably helped improve NUE over the past several decades in the United States and Japan (Cassman, Walters, and Dobermann 2002). For example, U.S. corn yield per amount of applied nitrogen has increased 36 percent over a period of about 20 years (Dobermann and Cassman 2005).

Unfortunately, these relatively simple practices are not enough to fine-tune fertilizer application to the nitrogen needs of a crop. This is partly due to the fact that different soils contain different

amounts of indigenous nitrogen, and partly because growing conditions and crop varieties alter a crop's response to applied nitrogen (Cassman, Walters, and Dobermann 2002).

More effective synchronization between crop growth and the amount and timing of nitrogen application therefore requires the calibration of indigenous soil nitrogen, crop variety, weather, and other factors that may affect growth rates. Measurements of soil nitrogen show considerable variation, even on a scale as small as a few meters. Ideally, many closely spaced measurements are needed to apply fertilizer with great precision, but as a substitute for such large numbers of measurements, researchers have attempted to adjust nitrogen applications on a similarly fine scale by using remote sensing of crop growth characteristics that respond to soil nitrogen availability. For example, variable fertilizer application rates have been adjusted on a per-meter basis by using tractor-mounted sensors that measure light reflectance from plant leaves (Raun et al. 2002). Although some improvements in NUE have been demonstrated in experiments using such methods, nitrogen measurements must be calibrated for each location.

Given the technology requirements—including GPS systems and remote crop sensors linked to fertilizer applicators—these high-precision methods may be more applicable to large farms in wealthy

nations (Weibe and Gollehon 2006b) than those in developing countries, or smaller farms generally.

Adoption by U.S. farmers of yield monitors used to adjust nitrogen applications reached 36.5 percent for corn in 2001, and 28.7 percent for soybeans in 2002. But only one-third of those farmers (or fewer) have also adopted yield-mapping of fields or high-precision, variable-rate fertilizer applicators. The use of such applicators fell from 12.3 percent for corn in 1998 to 9.8 percent in 2001, and from 6.7 percent for soybeans in 1998 to 5.0 percent in 2002 (Weibe and Gollehon 2006b). This suggests that farmers have shown resistance to adopting more advanced forms of precision agriculture.

For soils with low levels of organic matter and, therefore, indigenous nitrogen, larger amounts of added fertilizer are required to meet yield goals. This will make it more difficult to achieve high levels of NUE. Because precision farming does not address the problem of poor-quality soils—especially soils with low or declining organic matter—it seems unlikely that this technique can address the problem of nitrogen pollution by itself. Nevertheless, the available data suggest that the use of precision farming where appropriate, along with organic farming and the use of cover crops that increase soil organic matter and indigenous nitrogen over time, should be encouraged to help meet NUE goals.

## CHAPTER 6

### *Conclusions and Recommendations*

---

**T**he impact of reactive nitrogen pollution on our air, water, and climate demands that we make better use of this invaluable resource. At the same time, our growing global population means that we also need to produce more food in the coming decades—a process that will worsen nitrogen pollution unless we change our current practices.

A single approach to improving NUE is not likely to reverse the current environmental degradation caused by industrial agriculture. Instead, we need to work toward the simultaneous improvement of crops, fertilizer usage, and, especially, methods that increase soil organic matter and indigenous nitrogen.

So far, GE has not produced commercially viable crops with physiologically complex traits such as improved NUE.<sup>12</sup> Although a few genes that appear promising for improving NUE have been identified in the public literature, they have yet to demonstrate that they can improve NUE consistently in various environments, and without significant undesirable side effects that could harm our agriculture, environment, or public health. In addition, the NUE values initially reported for several of these genes must be considered preliminary, because most of the tests were not conducted in the environment over an extended period of time.

The single non-GE crop varieties that have been used to gauge the NUE of GE varieties are not sufficient to determine the degree to which an engineered gene may improve NUE compared with available varieties of the crop. It is possible, for example, that a GE variety may have lower

NUE than one or more commercial crop varieties against which it has not been compared. And because much of the testing of GE crops is conducted behind closed doors, public assessment of the efficacy and safety of these crops will have to await their emergence from the regulatory process.

#### **The Promise and Pitfalls of Non-GE Approaches**

Traditional breeding has improved both NUE and crop yields over the past several decades (Table 2), and it seems likely that it can continue to help improve NUE in coming years. Current evidence does not show that GE has any clear advantages over traditional breeding for improving NUE. In fact, the limited data available suggest that genetic variation for NUE within crop species may be as high as has been shown so far for engineered genes from other species.

Since little visible effort has been made thus far to explore this variation, either within crop species or their sexually compatible wild relatives, the potential exists for improving NUE by making use of this variation through breeding. As with GE, however, it is possible that NUE traits within the crop gene pool could have unintended negative side effects. But we do not believe this risk is as high for genes that are part of the normal crop genome as it is for exotic genes introduced to the crop genome through GE, or engineered genes expressed in ways outside the typical range of crop metabolism.

NUE traits identified only as quantitative trait loci, which may be used in traditional breeding, face logistical challenges because of the possibility

---

<sup>12</sup> Current GE crops have been engineered simply to produce the desired GE protein, not to create a plant with a significantly different metabolism (as would be needed to increase NUE).

that they may respond to different environments or different crop varieties in undesirable ways. Overall, however, traditional breeding shows considerable early promise for improving NUE.

Organic farming and other low-external-input methods including the use of cover crops show considerable promise as well. These methods have the additional benefit of addressing several agricultural problems simultaneously. For example, increasing soil organic content—which includes both carbon and nitrogen—can improve NUE and water retention while reducing nitrogen pollution, erosion, and pesticide use. These practices have received far too little attention from the research community and farm policy makers.

Finally, precision farming, broadly defined, may have already contributed to some improvement in NUE over the past several decades. These methods may continue to improve NUE in developed countries, although more technologically complex and precise methods do not appear to have been widely adopted so far. It is less clear how much they have to offer to small farms, especially in developing countries. Also, because precision farming does not address the fundamental problem of soil health by improving soil organic content over time, there are significant limits on how far it can improve NUE, especially for poor-quality soils.

Precision farming has received considerable research attention, in part because it is generally compatible with current industrial agriculture processes. While it deserves continued attention, this should not come at the expense of other promising approaches such as breeding and organic farming.

Several of the methods for improving NUE discussed here are largely complementary, although GE and breeding largely overlap in their possible contributions; both may reduce the need for added nitrogen to achieve a desired yield. Organic and similar methods can also reduce the need for added nitrogen, especially synthetic nitrogen, by building soil organic content and indigenous nitrogen over time. Precision farming can better match

the amount of added synthetic nitrogen to what crops actually need. Currently, however, traditional breeding and organic or similar sustainable methods receive only meager amounts of public research support and incentives.

## What the United States Should Do

Given the current state of affairs, the Union of Concerned Scientists offers the following recommendations:

- **Public crop breeding programs that include improved NUE as a goal should receive increased support.** This research should include evaluation of the genetic diversity available for improving NUE in the gene pools of crops and their compatible wild relatives.
- **Public breeding programs should be encouraged to develop crop varieties ready for commercial use,** in part so that alternatives to the GE NUE varieties emphasized by large seed companies are made available.
- **Organic and similar farming methods—especially the use of cover crops—should receive additional research support.** For example, the establishment and growth of legume cover crops should be improved, and new varieties and crops should be developed for various environments (such as colder climates). Research is also needed on the integration of cover crops into cash crop rotations, the use of mixtures of cover crops, and the efficacy of cover crops.
- **Crops should be developed for compatibility with organic and other sustainable methods** that can, for example, make the best use of indigenous nitrogen (and other nutrients) and organic nitrogen sources.
- **Developing countries and their farmers should be compensated for genetic resources** used by breeders in other countries through a meaningful consultation process.
- **Better methods are needed to identify, and new rules are needed to regulate, unintended side effects of GE.**

As noted previously (Gurian-Sherman 2009), the current regulations are inadequate.

- **Better data are needed on the measurement, efficacy, costs, and benefits or drawbacks of the various methods for improving NUE.** Organic and similar methods that are subsequently found to work well and provide multiple benefits should be supported with incentives.

As we have shown, the opportunities to address the problems caused by the overuse of synthetic nitrogen in agriculture are considerable. But achieving the degree of improvement in NUE

needed over the coming years will require increased public investment and a commitment to move beyond our current fixation on industrial agriculture methods such as precision farming and GE. We must begin providing more support for methods that have the greatest promise for the greatest good—that is, expanding our food supply while reducing the damage caused by nitrogen pollution.

## References

---

- Ameziane, R., K. Bernhard, and D. Lightfoot. 2000. Expression of the bacterial *gdhA* gene encoding a NADPH glutamate dehydrogenase in tobacco affects plant growth and development. *Plant and Soil* 221:47–57.
- Anderson, N., R. Strader, and C. Davidson. 2003. Airborne reduced nitrogen: Ammonia emissions from agriculture and other sources. *Environment International* 29:277–286.
- Animal and Plant Health Inspection Service. 2009a. Field test releases in the U.S. Information Systems for Biotechnology. Online at <http://www.isb.vt.edu/cfdocs/fieldtests1.cfm>.
- Animal and Plant Health Inspection Service. 2009b. Petitions of nonregulated status granted or pending by APHIS as of September 17, 2009. Online at [http://www.aphis.usda.gov/brs/not\\_reg.html](http://www.aphis.usda.gov/brs/not_reg.html).
- Badgley, C., J. Moghtader, E. Quintero, E. Zakem, M.J. Chappell, K. Avilés-Vázquez, A. Samulon, and I. Perfecto. 2007. Organic agriculture and the global food supply. *Renewable Agriculture and Food Systems* 22:86–108.
- Beatty, P.H., A.K. Shrawat, R.T. Carroll, T. Zhu, and A.G. Good. 2009. Transcriptome analysis of nitrogen-efficient rice over-expressing alanine aminotransferase. *Plant Biotechnology Journal* 7:1–15.
- Bernardo, R. 2008. Molecular markers and selection for complex traits in plants: Learning from the last 20 years. *Crop Science* 48:1649–1664.
- Brancourt-Hulmel, M., G. Doussinault, C. Lecomte, P. Be´rard, B. Le Buanec, and M. Trottet. 2003. Genetic improvement of agronomic traits of winter wheat cultivars released in France from 1946 to 1992. *Crop Science* 43:37–45.
- Cassman, K.G., A. Dobermann, and D.T. Walters. 2002. Agroecosystems, nitrogen-use efficiency and nitrogen management. *Ambio* 31:132–140.
- Coque, M., and A. Galais. 2006. Genomic regions involved in response to grain yield selection at high and low nitrogen fertilization in maize. *Theoretical and Applied Genetics* 112:1205–1220.
- Craine, J., W. Bond, W.G. Lee, P.B. Reich, and S. Ollinger. 2003. The resource economics of chemical and structural defenses across nitrogen supply gradients. *Oecologia* 142:547–556.
- Datta, K., R. Velazhahan, N. Oliva, I. Ona, T. Mew, G.S. Khush, S. Muthukrishnan, and S.K. Datta. 1999. Over-expression of the cloned rice thaumatin-like protein (PR-5) gene in transgenic rice plants enhances environmental friendly resistance to *Rhizoctonia solani* causing sheath blight disease. *Theoretical and Applied Genetics* 98:1138–1145.
- Dekkers, J.C.M., and F. Hospital. 2002. The use of molecular genetics in the improvement of agricultural populations. *Nature Reviews Genetics* 3:22–32.
- Diaz, R.J., and R. Rosenberg. 2008. Spreading dead zones and consequences for marine ecosystems. *Science* 321:926–929.

- Dobermann, A., and K.G. Cassman. 2005. Cereal area and nitrogen use efficiency are drivers of future nitrogen fertilizer consumption. *Science in China Series C: Life Sciences* 48:745–758.
- Drinkwater, L.E., P. Wagoner, and M. Sarrantonio. 1998. Legume-based cropping systems have reduced carbon and nitrogen losses. *Nature* 396:262–265.
- Duvick, D.N. 2005. The contribution of breeding to yield advances in maize (*Zea mays* L.). *Advances in Agronomy* 86:83–145.
- Ellstrand, N.C. 2003. *Dangerous liaisons? When cultivated plants mate with their wild relatives*. Baltimore and London: The Johns Hopkins University Press.
- Environmental Protection Agency (EPA). 2009a. *Nitrous oxide*. Washington, DC. Online at <http://www.epa.gov/nitrousoxide/sources.html>.
- Environmental Protection Agency (EPA). 2009b. *U.S. greenhouse gas inventory report: Agriculture*. Washington, DC. Online at <http://www.epa.gov/climatechange/emissions/downloads09/Agriculture.pdf>.
- Environmental Protection Agency (EPA). 2008. *Hypoxia in the northern Gulf of Mexico: An update by the EPA science advisory board*. EPA-SAB-08-003. Washington, DC.
- Fan, A.M., and V.E. Steinberg. 1996. Health implications of nitrate and nitrite in drinking water: An update on methemoglobinemia occurrence and reproductive and developmental toxicity. *Regulatory Toxicology and Pharmacology* 23:35–43.
- Galais, A., and B. Hirel. 2004. An approach to the genetics of nitrogen use efficiency in maize. *Journal of Experimental Botany* 55:295–306.
- Galloway, J.N., J.D. Aber, J.W. Erisman, S.P. Seitzinger, R.W. Howarth, E.B. Cowling, and B.J. Cosby. 2003. The nitrogen cascade. *Bioscience* 53(4):341–356.
- Galloway, J.N., E.B. Cowling, S.J. Seitzinger, and R. Socolow. 2002. Reactive nitrogen: Too much of a good thing? *Ambio* 31:60–63.
- Gepts, P. 2002. A comparison between crop domestication, classical plant breeding, and genetic engineering. *Crop Science* 42:1780–1790.
- Good, A.G., S.J. Johnson, M. De Pauw, R.T. Carroll, N. Savidov, J. Vidmar, Z. Lu, G. Taylor, and V. Stroehrer. 2007. Engineering nitrogen use efficiency with alanine aminotransferase. *Canadian Journal of Botany* 85(3):252–262.
- Good, A.G., A.K. Shrawat, and D.G. Muench. 2004. Can less yield more? Is reducing nutrient input into the environment compatible with maintaining crop production? *Trends in Plant Science* doi:10.1016/j.tplants.2004.10.008, 1–9.
- Goodman, M.M. 2004. Plant breeding requirements for applied molecular biology. *Crop Science* 44:1913–1914.
- Gurian-Sherman, D. 2009. *Failure to yield: Evaluating the performance of genetically engineered crops*. Cambridge, MA: Union of Concerned Scientists.
- Habash, D.Z., S. Bernard, J. Schondelmaier, J. Weyen, and S.A. Quarrie. 2007. The genetics of nitrogen use in hexaploid wheat: N utilisation, development and yield. *Theoretical and Applied Genetics* 114:403–419.
- Habash, D.Z., A.J. Massiah, H.L. Rong, R.M. Wallsgrove, and R.A. Leigh. 2001. The role of cytosolic glutamine synthetase in wheat. *Annals of Applied Botany* 138:83–89.

- Heffner, E.L., M.E. Sorrells, and J.-L. Jannink. 2009. Genomic selection for crop improvement. *Crop Science* 49:1–12.
- Hirel, B., J. Le Gouis, B. Ney, and A. Gallais. 2007. The challenge of improving nitrogen use efficiency in crop plants: Towards a more central role for genetic variability and quantitative genetics within integrated approaches. *Journal of Experimental Botany* 58:2369–2387.
- Hoisington, D., M. Khairallah, T. Reeves, J.-M. Ribaut, B. Skovmand, S. Taba, and M. Warburton. 1999. Plant genetic resources: What can they contribute toward increased crop productivity? *Proceedings of the National Academy of Sciences U.S.A.* 96:5937–5943.
- Kloppenborg, J.R., Jr. 2005. *First the seed: The political economy of plant biotechnology*. Second edition. Madison, WI: University of Wisconsin Press.
- Krupa, S.V. 2003. Effects of atmospheric ammonia (NH<sub>3</sub>) on terrestrial vegetation: A review. *Environmental Pollution* 124:179–221.
- Lam, H.-M., P. Wong, H.-K. Chan, K.-M. Yam, L. Chen, C.-M. Chow, and G.M. Coruzzi. 2003. Overexpression of the *ASN1* gene enhances nitrogen status in seeds of *Arabidopsis*. *Plant Physiology* 132:926–935.
- Lawlor, D.W. 2002. Carbon and nitrogen assimilation in relation to yield: Mechanisms are the key to understanding production systems. *Journal of Experimental Botany* 53:773–787.
- Letter, D.W., R. Siedel, and W. Liebhardt. 2003. The performance of organic and conventional cropping systems in an extreme climate year. *American Journal of Alternative Agriculture* 18(3):146–154.
- Long, S.P., X.-G. Zhu, S.L. Naidu, and D.R. Ort. 2006. Can improvement in photosynthesis increase crop yields? *Plant, Cell and Environment* 29:315–330.
- Marriott, E.E., and M.M. Wander. 2006. Total and labile soil organic matter in organic and conventional farming systems. *Soil Society of America Journal* 70:950–959.
- Martin, A., J. Lee, T. Kichey, D. Gerentes, M. Zivy, C. Tatout, F. Dubois, T. Balliau, B. Valot, M. Davanture, T. Tercé-Laforgue, I. Quilleré, M. Coque, A. Gallais, M.-B. Gonzalez-Moro, L. Bethencourt, D.Z. Habash, P.J. Lea, A. Charcosset, P. Perez, A. Murigneux, H. Sakakibara, K.J. Edwards, and B. Hirel. 2006. Two cytosolic glutamine synthetase isoforms of maize are specifically involved in the control of grain production. *The Plant Cell* 18:3252–3274.
- McCubbin, D.R., B.J. Apelberg, S. Roe, and F. Divita, Jr. 2002. Livestock ammonia management and particulate-related health benefits. *Environmental Science and Technology* 36(6):1141–1146.
- Mickelson, S., D. See, F.D. Meyer, J.P. Garner, C.R. Foster, T.K. Blake, and A.M. Fischer. 2003. Mapping of QTL associated with nitrogen storage and remobilization in barley (*Hordeum vulgare* L.) leaves. *Journal of Experimental Botany* 54:801–812.
- Moll, R.H., E.J. Kamprath, and W.A. Jackson. 1987. Development of nitrogen efficient prolific hybrids of maize. *Crop Science* 27:181–186.
- Mungur, R., A.D.M. Glass, D.B. Goodenow, and D.A. Lightfoot. 2005. Metabolite fingerprinting in transgenic *Nicotiana tabacum* altered by the *Escherichia coli* glutamate dehydrogenase gene. *Journal of Biomedicine and Biotechnology* 5:198–214.

- Muurinen, S., G.A. Slafer, and P. Peltonen-Sainio. 2006. Breeding effects on nitrogen use efficiency of spring cereals under northern conditions. *Crop Science* 46:561–568.
- National Research Council. 2004. *Safety of genetically engineered foods: Approaches to assessing unintended health effects*. Washington, DC: National Academy Press.
- National Research Council. 2000. *Genetically modified pest-protected plants: Science and regulation*. Washington, DC: National Academy Press.
- Obara, M., M. Kajiuura, Y. Fukuta, M. Yano, M. Hayashi, T. Yamaya, and T. Sato. 2001. Mapping QTLs of cytosolic glutamine synthetase (GS1) and NADH-glutamate synthase in rice (*Oryza sativa* L.). *Journal of Experimental Botany* 52:1209–1217.
- Ortiz-Monesterio, J.I., K.D. Sayre, S. Rajaram, and M. McMahon. 1997. Genetic progress in wheat yield and nitrogen use efficiency under four nitrogen rates. *Crop Science* 37:898–904.
- Pimentel, D., P. Hepperly, J. Hanson, D. Douds, and R. Seidel. 2005. Environmental, energetic and economic comparisons of organic and conventional farming systems. *BioScience* 55:573–582.
- Raun, W.R., and G.V. Johnson. 1999. Improving nitrogen use efficiency for cereal production. *Agronomy Journal* 91:357–363.
- Raun, W.R., J.B. Solie, G.V. Johnson, M.L. Stone, R.W. Mullen, K.W. Freeman, W.E. Thomason, and E.V. Lukina. 2002. Improving nitrogen use efficiency in cereal grain production with optical sensing and variable rate application. *Agronomy Journal* 94:815–820.
- Ravishankara, A.R., J.S. Daniel, and R.W. Portmann. 2009. Nitrous oxide (N<sub>2</sub>O): The dominant ozone-depleting substance emitted in the 21st century. *Science Express* doi:10.1126/science.1176985.
- Rockström, J., W. Steffen, K. Noone, Å. Persson, F.S. Chapin III, E.F. Lambin, T.M. Lenton, M. Scheffer, C. Folke, H.J. Schellnhuber, B. Nykvist, C.A. de Wit, T. Hughes, S. van der Leeuw, H. Rodhe, S. Sörlin, P.K. Snyder, R. Costanza, U. Svedin, M. Falkenmark, L. Karlberg, R.W. Corell, V.J. Fabry, J. Hansen, B. Walker, D. Liverman, K. Richardson, P. Crutzen, and J.A. Foley. 2009. A safe operating space for humanity. *Nature* 461:472–475.
- Sanchez, P.A. 2002. Soil fertility and hunger in Africa. *Science* 295:219–220.
- Shrawat, A.K., R.T. Carroll, M. DePauw, G.J. Taylor, and A.G. Good. 2008. Genetic engineering of improved nitrogen use efficiency in rice by the tissue-specific expression of alanine aminotransferase. *Plant Biotechnology Journal* 6: 722–732.
- Smil, V. 2000. *Feeding the world: A challenge for the twenty-first century*. Cambridge, MA: MIT Press.
- Snapp, S.S., S.M. Swinton, R. Labarta, D. Mutch, J.R. Black, R. Leep, J. Nyiraneza, and K. O’Neil. 2005. Evaluating cover crops for benefits, costs, and performance within cropping system niches. *Agronomy Journal* 97:322–332.
- Strock, J.S., P.M. Porter, and M.P. Russelle. 2004. Cover cropping to reduce nitrate loss through subsurface drainage in the northern U.S. Corn Belt. *Journal of Environmental Quality* 33:1010–1016.

- Sweetlove, L.J., D. Fell, and A.R. Fernie. 2008. Getting to grips with the plant metabolic network. *Biochemical Journal* 409:27–41.
- Teasdale, J.R., C.B. Coffman, and R.W. Mangum. 2007. Potential long-term benefits of no-tillage and organic cropping systems for grain production and soil improvement. *Agronomy Journal* 99:1297–1305.
- Tonitto, C., M.B. David, and L.E. Drinkwater. 2006. Replacing bare fallows with cover crops in fertilizer-intensive cropping systems: A meta-analysis of crop yield and N dynamics. *Agriculture, Ecosystems and Environment* 112:58–72.
- Vitousek, P.M., J.D. Aber, R.W. Howarth, G.E. Likens, P.A. Matson, D.W. Schindler, W.H. Schlesinger, and D.G. Tilman. 1997. Human alteration of the global nitrogen cycle: Sources and consequences. *Ecological Applications* 7(3):737–750.
- Vitousek, P., S. Hattenschwiler, L. Olander, and S. Allison. 2002. Nitrogen and nature. *Ambio* 31:97–101.
- Vitousek P.M., R. Naylor, T. Crews, M.B. David, L.E. Drinkwater, E. Holland, P.J. Johnes, J. Katzenberger, L.A. Martinelli, P.A. Matson, G. Nziguheba, D. Ojima, C.A. Palm, G.P. Robertson, P.A. Sanchez, A.R. Townsend, and F.S. Zhang. 2009. Nutrient imbalances in agricultural development. *Science* 324:1519–1520.
- Wiebe, K., and N. Gollehon. 2006a. Nutrient management. In: Agricultural resources and environmental indicators. Economic Information Bulletin EIB-16. Economic Research Service, U.S. Department of Agriculture.
- Wiebe, K., and N. Gollehon. 2006b. Information technology management. In: Agricultural resources and environmental indicators. Economic Information Bulletin EIB-16. Economic Research Service, U.S. Department of Agriculture.
- Xu, K., X. Xu, T. Fukao, P. Canlas, R. Maghirang-Rodriguez, S. Heuer, A.M. Ismail, J. Bailey-Serres, P.C. Ronald, and D.J. Mackill. 2006. *Sub1A* is an ethylene-response-factor-like gene that confers submergence tolerance to rice. *Nature* 442: 705–708.
- Yamaya, T., M. Obara, H. Nakajima, S. Sasaki, T. Hayakawa, and T. Sato. 2002. Genetic manipulation and quantitative-trait loci mapping for nitrogen recycling in rice. *Journal of Experimental Botany* 53:917–925.
- Yanagisawa, S., A. Akiyama, H. Kisaka, H. Uchimiya, and T. Miwa. 2004. Metabolic engineering with *Dof1* transcription factor in plants: Improved nitrogen assimilation and growth under low-nitrogen conditions. *Proceedings of the National Academy of Sciences U.S.A.* 101: 7833–7838.

# NO SURE FIX

## Prospects for Reducing Nitrogen Fertilizer Pollution through Genetic Engineering



**A**gricultural operations currently apply massive amounts of synthetic nitrogen fertilizer to crops—more than what the plants can actually use. Much of the excess nitrogen escapes from the farm and becomes a major component of global pollution, contributing to global warming, acid rain, and “dead zones” in the ocean.

Genetic engineering (GE) that would enable crops to use nitrogen more efficiently has been proposed as a way of reducing nitrogen pollution while maintaining or increasing the productivity needed to feed an increasing global population. However, in *No Sure Fix*, the Union of Concerned Scientists finds that GE has yet to produce any crops capable of achieving this goal,

despite increasing research efforts over the past decade. Preliminary results for several genes show some promise, but the prospects for their commercial use are uncertain due to the complexity of nitrogen metabolism and genetics in crops.

Meanwhile, traditional plant breeding and other methods have shown success in increasing crops’ nitrogen use efficiency, but are currently neglected compared with GE. Reducing nitrogen pollution from agriculture while increasing crop yields is a challenge that will require increased support for multiple, complementary approaches, including traditional breeding, cover crops, and precision farming.

### National Headquarters

Two Brattle Square  
Cambridge, MA 02238-9105  
Phone: (617) 547-5552  
Fax: (617) 864-9405

### West Coast Office

2397 Shattuck Ave., Ste. 203  
Berkeley, CA 94704-1567  
Phone: (510) 843-1872  
Fax: (510) 843-3785

### Washington, DC, Office

1825 K St. NW, Ste. 800  
Washington, DC 20006-1232  
Phone: (202) 223-6133  
Fax: (202) 223-6162

### Midwest Office

One N. LaSalle St., Ste. 1904  
Chicago, IL 60602-4064  
Phone: (312) 578-1750  
Fax: (312) 578-1751

**Web:** [www.ucsusa.org](http://www.ucsusa.org)

**Email:** [ucs@ucsusa.org](mailto:ucs@ucsusa.org)



Union of  
Concerned  
Scientists

Citizens and Scientists for Environmental Solutions