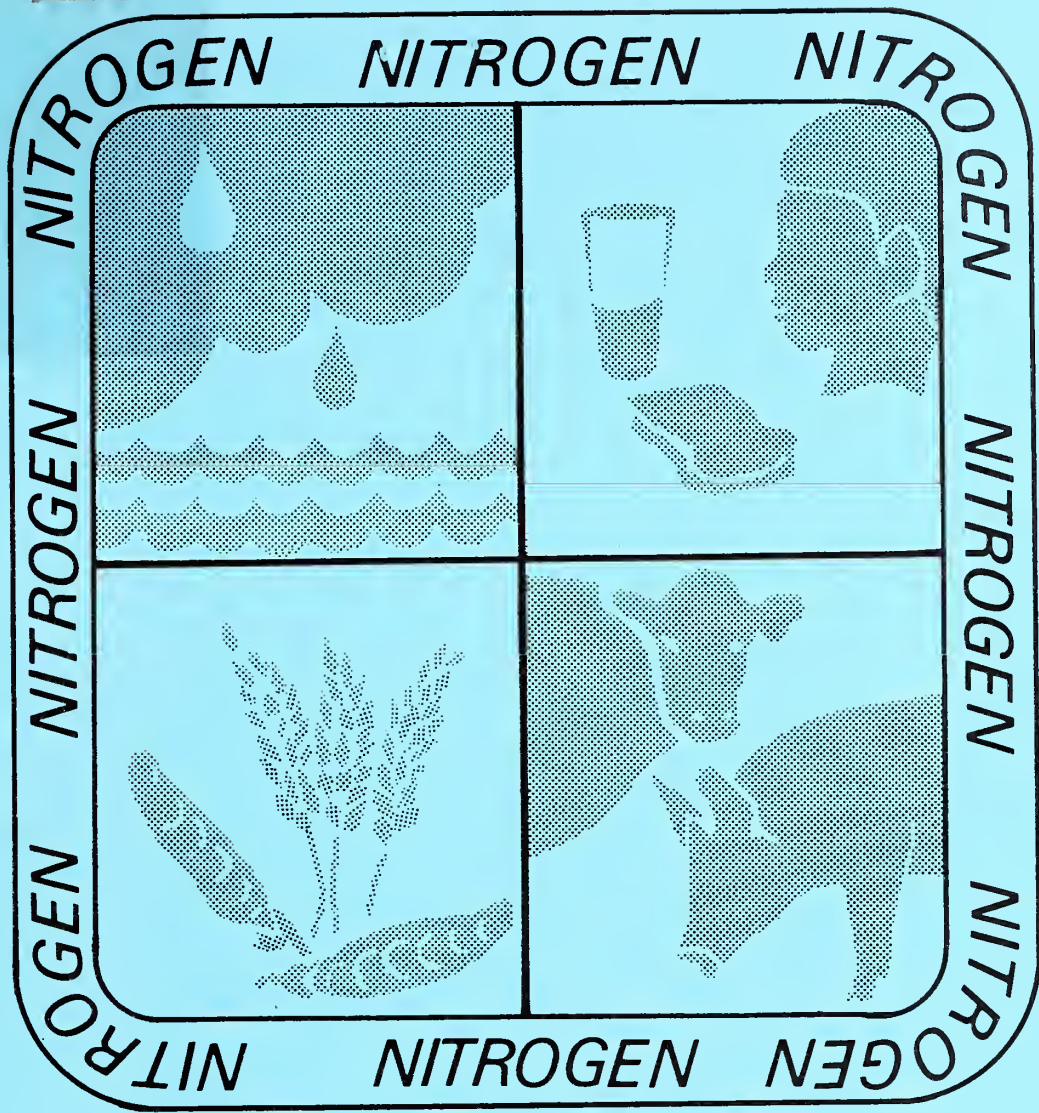


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# NITROGEN RESEARCH-1989

## Current Advances And Future Priorities

Agricultural Research Service  
United States Department of Agriculture

VF - NITROGEN

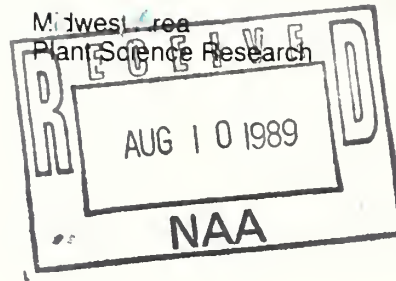
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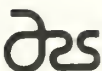
Dear Colleague:

We enclose a copy of the technical report for the USDA-ARS Working Conference on Nitrogen Research that was held in May 1989. The report charts progress in nitrogen-related research since the last review in 1977, identifies scientific and technological contributions, and suggests research imperatives and tasks that are relevant to major issues confronting U. S. society.

We trust that this information will be useful in planning and executing your research, development, and technology transfer programs.

Sincerely yours,

The Organizing Committee  
G. H. Heichel, Chair  
R. F. Follett  
J. F. Power  
D. R. Keister



Agricultural  
Research  
Service

United States  
Department of  
Agriculture

U.S.D.A., NAL  
Cataloging Prep

**NITROGEN RESEARCH 1989**

**Current Advances and  
Future Priorities**

**Technical Report of a USDA-Agricultural Research Service**

**Working Conference Held May 23-25, 1989**

**St. Louis, Missouri**



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## PREFACE

Nitrogen is a natural resource. It comprises 80% of the earth's atmosphere. The vast reserve of gaseous nitrogen is nutritionally worthless to plants, and to people and other animals, until it is transformed by either of two main processes into chemically active forms that can be used to build life-sustaining protein. One process occurs naturally, aided by unique bacteria living either in partnership with certain plants, or free in the soil. The second is an industrial process used to combine atmospheric nitrogen into manufactured fertilizers.

Chemically active nitrogen forms (e.g., ammonium, nitrate, amino acids, and protein) are essential for all living organisms. Without these forms, plants could not grow, and people and other animals could not breathe, eat, or reproduce. Life could not exist.

Although chemically active nitrogen forms support living creatures, they eventually return to the inert gaseous reserve in the atmosphere. In contrast to the two main pathways for the conversion of atmospheric nitrogen into chemically active forms, many natural and complex pathways return active nitrogen forms to the atmosphere.

In the course of return to the atmosphere, active nitrogen forms may temporarily elude their useful purposes and cause problems for society. Enrichment of streams and lakes with nitrate and ammonium reduces the recreational quality of water. Enrichment of drinking water with nitrate reduces its quality for human and livestock consumption. Losses of fertilizer forms of nitrogen reduce the profitability of farming. Losses of soil forms of nitrogen reduce the sustainability of farmland. Some gaseous forms of nitrogen, not yet fully inactive, contribute to the greenhouse effect, i.e., global climate change.

The natural cycle of providing nitrogen to support life, and returning it at life's end to the atmospheric reserve, is difficult to control; it leaks.

To reduce the undesirable effects of leakage of active nitrogen forms into the environment, humankind must intervene with wisdom gained from science and with knowledge gained from experience. Responsible stewardship of the nitrogen resource is essential. Public policy, regulations, and scientific inquiry are the bases of responsible stewardship. These functions are shared by many institutions and organizations, but the Agricultural Research Service of USDA, and its collaborators, have the lead role in science.

This document reports scientific contributions of the Agricultural Research Service over the past 12 years in enhancing the stewardship of the nitrogen resource. Advances in converting inactive atmospheric nitrogen to the chemically active forms essential to the nutrition of plants and people have been made. Opportunities to reduce the leakiness of the natural nitrogen conversion processes have been discovered.

This document also reports scientific objectives which, once enabled by public policy, will lead to improved stewardship of nitrogen, and contribute to better water quality, safer and sustainable methods of producing food, and reduced risk of undesirable climatic change.



## ACKNOWLEDGMENTS

The need for a workshop on one component of nitrogen research, biological nitrogen fixation, was first articulated by E.B. Knipling. J.P. Miksche proposed increasing the scope of a workshop to integrate coverage of the molecular, organismal, and ecosystem issues in nitrogen research. The workshop became a Working Conference with 76 participants.

Several individuals deserve special recognition for facilitating this Conference. Jan Overton, St. Paul, executed the required keyword search of AD-416s and AD-421s. Dennis Campion, Peoria, agreed to MWA support for the Conference. Rollin Lawrence, Peoria, arranged for facilities in St. Louis, and offered the services of his secretary, Beverly Snyder, to provide word processing support for the Conference; her help was indispensable. Marilyn Hole, St. Paul, typed much of the final report except for the white papers, which were printed as received. Kristen Kirkeby, St. Paul, designed the cover.

We thank the participants in the Conference, whose enthusiasm and contributions made it a success.

The Organizing Committee and Editors

G.H. Heichel  
R.F. Follett  
J.F. Power  
D.R. Keister

August, 1989



## EXECUTIVE SUMMARY

Seventy-six federal, state, and private sector agricultural scientists met in St. Louis, MO, May 23-25, 1989, to conduct the first comprehensive assessment of the USDA-Agricultural Research Service program in nitrogen research held since 1977. The Conference was structured around contemporary societal interests in agricultural sustainability, water quality, and global climate change. Keynote speakers and comprehensive review papers increased the awareness of participants to the current status of science in nitrogen research, and to the need to target science at socially-important problems.

The structured program provided an assessment of technological and scientific contributions from nitrogen research over the past 12 years, and an assessment of socially-important and scientifically-sound research imperatives and tasks for the future. Spontaneous discussion and deliberation among participants revealed concern about the roles and responsibilities of ARS scientists in technology transfer.

### Contributions from Nitrogen Research.

The relevance of an agricultural research program to social needs can be gauged by its technological ("science put to practice") and scientific ("advancing the frontiers of knowledge") contributions. This is especially important for the program in nitrogen research, which has been very active since the last comprehensive review in 1977. In the brief time scheduled, participants identified 26 major technological and 38 major scientific contributions that emerged from the nitrogen program in recent years. These relate to:

#### \* Sustainability of Agriculture

- 13 technological advances encompassing development of new crop germplasm, development of new or improved management practices, and patenting of bacterial and plant technologies.
- 19 scientific advances in plant and bacterial genetics, metabolism, molecular biology, and soil and crop management.

#### \* Water Quality

- 12 technological advances encompassing modeling, soil testing, irrigation practices, and crop and animal residue management.
- 11 scientific advances on developing analytical methods, determining soil and water nitrogen transformations, improving management practices, and developing simulation models.

#### \* Global Climate Change

- Technological developments in modeling plant responses to environmental stresses.
- 8 scientific advances encompassing modeling and the losses of gaseous nitrogen forms to the atmosphere.

## Research Opportunities.

The most socially and scientifically important research ideas were arrayed as 24 tasks within four comprehensive imperatives:

### Imperative 1. Develop and Use Plant Germplasm with Improved Attributes.

#### Research Tasks

- a) Identify and map plant and bacterial genes controlling uptake, assimilation, partitioning, and utilization of N ( $N_2$ ,  $NO_3^-$ ,  $NH_4^+$ ).
- b) Determine the physiological, biochemical, and molecular regulation of N ( $N_2$ ,  $NO_3^-$ ,  $NH_4^+$ ) assimilation and partitioning at the cellular, organismal, and crop community levels.
- c) Improve host compatibility and capability for  $N_2$  fixation of root nodule bacteria.
- d) Identify germplasm with improved capability for  $N_2$  fixation and soil N use efficiency, and develop strategies to use the new germplasm in plant improvement programs.
- e) Understand the interdependence of plants and rhizosphere organisms such as *Pseudomonas* spp. and VA mycorrhizae, and their interaction with root nodule bacteria.
- f) Determine effects of environmental stresses, especially those anticipated from global climate change, on plant N metabolism.
- g) Develop legume cultivars with special attributes, e.g., ability to reseed, winter-hardiness, efficient water use, and efficient N scavenging, for use as cover crops and intercrops in new cropping systems.

### Imperative 2. Increase the Understanding of Soil N Transformation Processes.

#### Research Tasks

- a) Determine factors controlling mineralization-immobilization processes, and conceive suitable analytical techniques to allow development of management practices to synchronize soil N availability with crop needs.
- b) Devise analytical methods to measure denitrification, ammonia volatilization, and other gaseous N transformations and determine the factors controlling these processes.
- c) Improve analytical methods to measure nitrification, and determine how to control nitrification and non-exchangable N transformations by inhibitors and by other methods.
- d) Determine the factors that control the accumulation and decomposition of soil organic matter, and their effects upon the soil physical, chemical, and biological environment.

- e) Assess the dynamics and relative importance of gaseous N exchange between the biosphere and atmosphere, and its potential cumulative effects on global warming.

Imperative 3. Conceive, Develop, and Facilitate Adoption of Acceptable N Management Systems.

Research Tasks.

- a) Determine effects of tillage and crop residue management practices on soil environment and N availability, and on subsequent N transformations and fate.
- b) Evaluate how cropping systems (including cover crops) cause changes in the soil environment and subsequent transformations and fate of N.
- c) Understand how soil and fertilizer management practices cause changes in the soil environment and subsequent N transformations, crop uptake, and fate of N.
- d) Evaluate effects of irrigation and drainage systems on changes in soil environment and subsequent N transformations and fate of all N sources.
- e) Conceive and develop suitable soil and plant tissue tests to accurately assess N deficiencies and to determine fertilizer N requirements for various soils, cropping systems, and climates.
- f) Develop the means to accurately predict mineralization rate of N from crop residues, animal manures, and other organic sources when used with various soils, tillage practices, and climates.
- g) Develop management practices that synchronize the availability of N from all sources with crop N requirements to minimize N losses to atmosphere and water.

Imperative 4. Integrate and Transfer Knowledge by Simulation and Modeling.

Research Tasks

- a) Develop accurate, process-based algorithms that quantitatively describe the effects of soil, crop, and climate on various N transformations.
- b) Incorporate algorithms into computer simulation models to accurately describe the effects of changes in soil environment resulting from management decisions on the fate of N from all sources, and on crop N uptake.
- c) Develop user-friendly computer models suitable for routine use by clientele that outline the consequences of alternative soil and crop management decisions in terms of crop production, net income, soil quality changes, and environmental quality.
- d) Develop computer models incorporating the foregoing algorithms and on-board sensors to change field equipment settings for planting and fertilization as the equipment passes from one soil condition to

another in the field.

- e) Field verify the foregoing models and establish mechanisms for transferring the technology to producers, agri-chemical dealers, extension and action agencies, consultants, and other clientele.

### Technology Transfer: Issues and Opportunities

Conference participants were sensitive to the widely-held public perception that the results of ARS research are poorly linked with the mainstream needs of society. The perception seems to conflict not only with the facts, but also with the intent and purpose of the 1984 ARS Technology Transfer Plan. Although ARS scientists are being increasingly and forcefully reminded to be responsive to the needs of society, the participants think that formidable barriers to doing so exist because (a) the reward system for ARS scientists principally emphasizes serving scientific peers, (b) agency regulations and protocol tightly govern information exchange with national and state legislators, and (c) scientific linkages with clientele in other federal agencies are often inhibited by bureaucratic turfing.

Conference participants recommend that the Agency comprehensively (a) assess its contemporary expectations of ARS scientists in technology transfer, and (b) conceive and implement policies to allow these expectations to be met. Without being exhaustive, the following questions deserve attention:

- \* Should ARS research be planned with technology transfer as an objective?
- \* Should research programs and scientist position descriptions in which technology transfer activities are a condition of employment be developed within ARS?
- \* Should the ARS reward system recognize technology transfer to the larger public constituency as a legitimate scientific role?
- \* How can perceived and established bureaucratic barriers to interagency collaboration be best addressed and removed?
- \* Should ARS scientists be more involved in periodic conferences and briefing sessions on high visibility topics to report Agency activities to legislative staffers, media, public interest groups, producer groups, and other federal agency personnel?
- \* How can current ARS literature and scientific publications be more effectively transmitted to Federal and State action agencies for immediate use in addressing societal needs?



## INTRODUCTION

The goal of the organizing committee of this conference was to provide ARS scientists with an enriching experience, through thoughtful and informed deliberation, that would lead to the formulation of guidelines for future ARS N research. Science cannot operate in isolation, but neither can it effectively operate in a crisis environment. Scientists must address real world problems and to do so requires that they be well-informed. Information flow is facilitated when peers meet to exchange ideas and to identify potential collaborators from other disciplines and locations.

This Conference was the first such overview of ARS research on N since 1977. During this 12 year interval the needs of society have changed. The American people have become more aware of and concerned about issues relating to agriculture that may affect their lives and the well-being of the nation. Some of the issues identified that may relate to research conducted in ARS are the following:

- \* Conserving, preserving, and enhancing our natural resources (air, land, water, germplasm, and human capital).
- \* Developing plant and animal production systems that are sustainable and profitable (economically efficient).
- \* Controlling and reducing environmental hazards associated with agriculture (ground water depletion, surface- and ground-water contamination by nitrogen, nitric and nitrous oxides in the atmosphere).
- \* Determining the potential impacts of biogenic trace gases on tropospheric and stratospheric ozone chemistry as related to the possible role of agriculture.
- \* Decreasing the potential impacts of the changing price of energy as related to the cost of inputs for agricultural production.
- \* Meeting needs mandated by Congress and identified by regulatory or action agencies.
- \* Advancing scientific frontiers acknowledged by the larger scientific community to be important.
- \* Producing commodities competitive on world markets.

These issues must be addressed by ARS to fulfill its research role for the USDA.

The conference opened with two invited presentations: Dr. Francis Clark (an eminent retired scientist and member of the ARS Hall of Fame) presented a long-term examination of research and issues related to the 'leaky N cycle.' Dr. Charles Benbrook (Executive Director, Board on Agriculture of the National Research Council) spoke on 'Nitrogen problems in the real world: perceptions of reality.' These provided attendees with a heightened awareness of the historical and current perspective of research on N and the current environment in which policy decisions concerning agricultural research are made.

To define specific areas of N research of concern to ARS, 'white papers' were presented on the following topics:

Advances in Assessing and Managing N Losses to the Environment. S.J. Smith, J. Schepers, and L.K. Porter.

Advances in N Use Efficiency. A. Halvorson, B. Bock, and J.J. Meisinger.

Advances in N Self-Sufficiency of Plants. J. Harper, P. Cregan, and C.P. Vance.

Crop Management Systems for Efficient N Use. M.P. Russelle and W. Hargrove.

These white papers defined the problem, gave an historical perspective, summarized problems solved and scientific achievements of the past 10 to 15 years, and identified research needs for deliberation and amplification by working groups at this conference. The white papers are included in this report; the authors are especially commended for the extensive and thorough manner in which their assignments were completed.

Following the presentations and discussions of the 'white papers,' working groups were organized for the following six subject matter areas to discuss and identify research priorities on:

- \* Soil Nitrogen Transformations
- \* Fertilizer Management
- \* Organic Sources of Nitrogen
- \* Water Quality
- \* Losses of N to the Atmosphere
- \* Nitrogen Nutrition of plants

After defining and prioritizing research needs for each subject matter area, attendees ranked the research needs according to their relevance to the following five crop management systems:

- \* Livestock Systems
- \* Cash Grain Systems
- \* Horticulture- and Vegetable-Crop Systems
- \* Irrigated Cropping Systems
- \* Dryland Systems

Finally, research needs for each crop management system were evaluated for their relevance to one or more of the following societal issues:

- \* Sustainability of Agriculture
- \* Water Quality
- \* Climate Change

The results of these deliberations comprise the research imperatives and tasks listed in this report.

In closing, one purpose of this conference was to summarize research problems which have been solved as well as urgent science that yet remains to be done and to report this information to ARS administrators and other users. Another equally important purpose of this conference was to nurture a vital and active research program on N that is national in scope, focused upon high priority issues, and consistent with views of individual scientists in identifying the high priority research imperatives and tasks needed to solve urgent contemporary problems.

PROGRAM  
OF THE  
USDA-AGRICULTURAL RESEARCH SERVICE WORKING CONFERENCE

MAY 23-25, 1989

HENRY VIII HOTEL AND CONFERENCE CENTER  
ST. LOUIS, MISSOURI

"Nitrogen Research 1989: Current Advances and Future Priorities"

- Purposes:
- \* Determine problems solved and scientific achievements.
  - \* Identify urgent problems to solve and urgent science to do.
  - \* Provide scientist participants a perspective of national ARS missions and the individual's role in multidisciplinary attack.
  - \* Provide informative, enriching experience for all participants.
  - \* Communicate contributions and program purposes to users with summary documents.

## PROGRAM

### **MONDAY, May 22 Conference Office and Registration: Wellington Room, 1st Floor**

Afternoon and Evening: Arrival and Registration  
Henry VIII Hotel

### **TUESDAY, May 23 [Royal Sussex Ballroom, 2nd Floor]**

- 8:00 - 8:15 a.m. Introduction and Announcements
- 8:15 - 8:35 a.m. "Charge to the Participants"  
Dr. W.D. Kemper, NPL, USDA-ARS-NPS
- 8:35 - 8:45 a.m. "Expectations"  
Dr. Gary Heichel, USDA-ARS

#### Keynote Addresses:

- 8:45 - 9:05 a.m. "Examining the Leaky Nitrogen Cycle."  
Dr. Francis Clark, USDA-ARS (Retired)
- 9:05 - 9:45 p.m. "Nitrogen Problems in the Real World: Perceptions of  
reality."  
Dr. Charles Benbrook, Executive Director, Board on  
Agriculture, NRC.
- 9:45 - 10:00 p.m. Discussion
- 10:00 - 10:30 a.m. Break

#### WHITE PAPERS

- 10:30 - 11:15 a.m. "Advances in Assessing and Managing N Losses to the  
Environment."  
S.J. Smith, J. Schepers, and L.K. Porter
- 11:15 - 11:35 a.m. Discussion
- 11:35 - 1:00 p.m. Lunch (on your own)

#### White Papers, continued: [Sussex, 2nd Floor]

- 1:00 - 1:45 p.m. "Advances in N Use Efficiency."  
A. Halvorson, B. Bock, and J.J. Meisinger
- 1:45 - 2:05 p.m. Discussion
- 2:05 - 2:50 p.m. "Advances in N Self-Sufficiency of Plants."  
J. Harper, P. Cregan, and C.P. Vance
- 2:50 - 3:10 p.m. Discussion

- 3:10 - 3:30 p.m. Break
- 3:30 - 4:15 p.m. "Crop Management Systems for Efficient N Use."  
M.P. Russelle and W. Hargrove
- 4:15 - 4:35 p.m. Discussion
- 4:35 - 4:45 p.m. Announcements
- 4:45 - 7:00 p.m. Relaxation and Dinner (on your own)

Evening Forum [Sussex, 2nd Floor]

- 7:00 - 8:20 p.m. Reflect on the keynote addresses and white papers.

At the guidance of the moderator, discuss the anticipated effects on ARS N research programs of the following scenarios (in succession):

- a) High vs. low energy costs (20 min)
- b) Restricted vs unrestricted N fertilizer use (20 min)
- c) Limited vs unlimited acres in production (20 min)
- d) Major climate change (e.g. a severe drought) (20 min)

- 8:20 - 8:30 p.m. Announcements and Adjourn

**WEDNESDAY, May 24**

Working Group Sessions by Research Activity

- 8:00 - 8:15 a.m. Announcements and Instructions [Sussex, 2nd Floor]
- 8:15 - 9:45 a.m. Convene Working Groups (Assignments in Appendix I)

- 1. Soil Nitrogen Transformations [Regency, 1st Floor]
- 2. Fertilizer Management [Bedford, 1st Floor]
- 3. Organic Sources of Nitrogen [Oxford, 2nd Floor]
- 4. Water Quality [Tudor, 2nd Floor]
- 5. Losses of Nitrogen to Atmosphere [St. George, 2nd Floor]
- 6. Nitrogen Nutrition of Plants [King James, 2nd Floor]

Working Group Assignments

- a) Define scope and boundaries of the group topic. (20 min)
- b) Each participant briefly present research (or policy) interests (or issues) and ideas for future research. (70 min)

- 9:45 - 10:00 a.m. Break

10:00 - 12:00 noon Reconvene Working Groups

- c) Participants brainstorm past achievements (major problems solved) and future research imperatives (major problems to solve). (70 min)
- d) Group reach consensus on past achievements and future priorities. (20 min)
- e) Group chair and recorder outline written report for presentation to all participants. (30 min)

12:00 - 1:15 p.m. Lunch (on your own)

1:15 - 2:15 p.m. Report of forenoon working groups (10 min each)  
[Sussex, 2nd Floor]

#### Working Group Sessions by Crop Management System

2:15 - 2:30 p.m. Announcements and Instructions [Sussex, 2nd Floor]

2:30 - 3:45 p.m. Convene Working Groups (Assignments forthcoming)

- 1. Livestock Systems [Tudor, 2nd Floor]
- 2. Cash Grain Systems [St. George, 2nd Floor]
- 3. Horticulture-Vegetable Systems [Oxford, 2nd Floor]
- 4. Irrigated Cropping Systems [Beford, 1st Floor]
- 5. Dryland Systems [Regency, 1st Floor]

#### Working Group Assignments

- a) Quickly discuss future research imperatives from each forenoon working group by crop management system (6 groups x N imperatives - 6N ideas to rank within each system). (60 min)
- b) Consensus ranking of 6N research imperatives by crop management system. (15 min)

3:45 - 4:00 p.m. Break

4:00 - 6:00 p.m. Reconvene Working Groups

- c) Within each crop management system, consensus allocation and ranking of each of the 6N research imperatives from (b) within the following societal issues:

- \* Sustainable Agriculture
- \* Water Quality
- \* Climate Change

- d) Group chair and recorder outline written report for presentation to all participants. (20 min)

5:20 - 5:50 p.m. Reports of 5 afternoon working groups (6 min each)  
[Sussex, 2nd Floor] (Submit hard copy to Organizing Committee)

5:50 - 6:00 p.m. Announcements and Adjourn [Sussex, 2nd Floor]

6:00 p.m. Dinner (on your own) Evening Open

Chairs and recorders of forenoon Working Groups 1-6 and  
afternoon working Groups 1-5 prepare final reports for  
Thursday morning.

**THURSDAY, May 25 [Royal Sussex Ballroom, 2nd Floor]**

8:30 - 9:30 a.m. Final Reports of Forenoon Working Groups (1-6)  
(10 min each)

9:30 - 10:00 a.m. Discussion

10:00 - 10:30 a.m. Break

10:30 - 11:00 a.m. Final Reports of Afternoon working Groups (1-5)  
(12 min each)

11:00 - 11:30 a.m. Discussion

11:30 a.m. Closing Remarks and Adjournment

Lunch (on your own)

1:00 p.m. Organizing Committee and Working Group Chairs and  
Recorders finalize hard copy of all reports.  
[St. George, Tudor, Wellington as needed]

Departure

## APPENDIX I.

### ASSIGNMENTS TO FORENOON WORKING GROUPS 1-6, MAY 24

Organizing Committee: G. Heichel  
R. Follet  
D. Keister  
J. Power

Conference Secretary: Beverly Snyder  
USDA-ARS MWA Administrative Office, Peoria

#### 1. Soil N Transformations:

L. Elliott (Chair)	J. Reeder	J. Smith
L.K. Porter (Recorder)	M. Russelle	D. Stott
B. Meek	M. Shaffer	F.E. Clark
R. Swank	D. Francis	R. Hauck

#### 2. Fertilizer Management:

D. Karlen (Chair)	A. Halvorson	S.C. Rao	B. Bock
A. Bauer (Recorder)	D. Westerman	D. Tanaka	F. West
W. Berg	J. Meisinger	G. Varvel	
H. Eck	A. Olness	H. Reetz	

#### 3. Organic Sources:

S. Wilkinson (Chair)	W. Honeycutt	P. Hunt	D. Lauer
G. Schuman (Recorder)	R.H. Dowdy	J. Radke	J. Power
A. Black	T.H. Dao	P. Rasmussen	
R. Janke	W. Hargrove	M. Vigil	

#### 4. Water Quality:

J. Schepers (Chair)	R. Lowrance	R. Schnabel
W. Guenzi (Recorder)	L. Owens	B. Volk
D. Carter	S.J. Smith	B. Vining
R. Follett	F. Thicke	D. Farrell

#### 5. Losses to Atmosphere:

A. Mosier (Chair)	D. Timmons
J. Doran (Recorder)	W. Heck
G. Hutchinson	D. Glotfelty

#### 6. Nitrogen Nutrition of Plants:

C. Vance (Chair)	P. Cregan	T. Rufty	J. Newton
W. Newton (Recorder)	J. Harper	T. Kaneshiro	R. Linderman
C.T. MacKown	W. Hunter	D. Kuykendall	D. Keister
D.K. Barnes	D. Israel	T. Sinclair	G. Heichel



## APPENDIX II.

### ADDITIONAL UNASSIGNED PARTICIPANTS

These persons should arrive prepared to join 1 of 6 forenoon working groups, and 1 of 5 afternoon working groups on Wednesday, May 25.

1. USDA-ARS National Program Leaders

W.D. Kemper  
D. Farrell

2. Non-ARS Participants

a. US-EPA:

Tim Amsden, Director, Groundwater Protection, Kansas City  
Robert Swank, Environmental Research Laboratory, Athens

b. Extension Service:

Francis Thicke, Soils Specialist, Washington, D.C.

c. Tennessee Valley Authority:

Bert Bock, Soil Scientists, Muscle Shoals  
Roland Hauck, Soil Scientist, Muscle Shoals

d. Rodale Institute:

Rhonda Janke, Agronomist, Kutztown, PA

e. The Fertilizer Institute:

Ford West, Washington, D.C.

f. University of Georgia:

William Hargrove, Professor

g. Soil Conservation Service:

Barbara Vining, Lincoln, NE

h. University of Missouri:

Bob Volk, Chair, Department of Agronomy

i. Potash and Phosphate Institute:

Harold Reetz

j. University of Nebraska:

Dennis Francis

k. Kansas State University:

Merle Vigil

l. Board on Agriculture, National Research Council:

Charles Benbrook, Executive Director



WHITE PAPERS



## ADVANCES IN ASSESSING AND MANAGING NITROGEN LOSSES TO THE ENVIRONMENT

S. J. Smith, J. S. Schepers and L. K. Porter  
U. S. Department of Agriculture  
Agricultural Research Service

### ABSTRACT

Environmental concern about excessive agricultural nitrogen (N) losses stems from both water quality and atmospheric standpoints. Available soil N supplies are generally inadequate for optimum crop production, so N is expected to continue as the fertilizer most applied to agricultural land. Basically, the solution to minimizing the potential for agricultural N — environmental problems is to manage land so as to enhance soil and supplemental N — use efficiency. The treatment here considers advances made in assessing N losses to wind, leaching, surface runoff, and volatilization, and the application of fertilizer, cropping, tillage, and irrigation techniques to minimize these losses. Overall, an integrated, or holistic, N management approach is recommended, with a goal of maintaining acceptable crop production and minimizing N leakage to the environment. Considered, too, are research needs for achieving this goal, involving particulate, soluble, and gaseous N forms. In the case of soluble N, sandy soils, conservation tillage, tile drainage, and irrigation warrant special attention.

### INTRODUCTION

Major concern continues to exist about potentially harmful environmental impacts associated with excessive agricultural nitrogen (N) losses. Concern about the losses stems from both water quality and atmospheric standpoints (Gilliam et al., 1985). Water quality concern has focused mainly on ground-water nitrate levels considered health threatening and accelerated eutrophication of surface waters, whereas atmospheric concern has focused on increased emission of nitrous oxide associated with N fertilizer application. Concern about increased nitrous oxide emission stems from its implication in stratospheric reactions contributing to depletion of the ozone, which protects the earth from ultraviolet radiation.

Nitrogen, an essential, major plant nutrient, represents the mineral fertilizer most applied to agricultural lands. This is because available soil N supplies are generally inadequate for optimum crop production. In the ammonium form, N is fairly immobile in soil. However, under most conditions, ammonium is converted biologically to nitrate which readily moves with the soil water. That nitrate not utilized by the crop has the potential to move from the soil to ground water, streams, and impoundments. In addition, some nitrate may be denitrified and pass to the atmosphere in a gaseous form. Dissipation to the atmosphere is also possible with ammonium, especially under alkaline soil conditions. Moreover, considerable particulate soil organic nitrogen is moved due to erosional processes. Overall, then, agricultural nitrogen may be lost to the environment in liquid, gaseous, or solid forms.

This paper considers pertinent factors and approaches useful in assessing and managing agricultural N losses to the environment. The treatment here covers work primarily over the past 10—15 years. For earlier information, readers are referred to publications by Stanford et al. (1970), Viets and Hageman (1971) and Wadleigh (1968). Basically, however, the solution to minimizing agricultural N losses to the environment remains the same, that is, increasing fertilizer N efficiency. Not to be overlooked are the environmental benefits N fertilizers provide. By increasing crop cover, they can reduce runoff and soil erosion. In addition, N fertilizer application to the more productive lands allows less suitable, fragile lands to be removed from cultivation.

## CONTRIBUTIONS TO N INPUT

To place the topic in proper perspective, it is important to recognize that N from natural sources has always existed in soils and waters. This N has originated from geological, biological, and atmospheric sources. In fact, some soil extracts in California have been reported to contain as much as 2000 mg nitrate-N L<sup>-1</sup>, primarily due to geological sources (Strathouse et al, 1980). Moreover, it is well to note that soils in their virgin states contained considerably higher N contents than they do today, under cropped, fertilized conditions. For example, in a study involving 8 major U. S. agricultural soils (Smith and Young, 1975), total N contents of the virgin, level, surface horizons contained 760 to 4110 mg TKN kg<sup>-1</sup>, on the average, 1/3 more than their N fertilized, cultivated counterparts that had been farmed 15 to 70+ years. Consequently, from an environmental standpoint, the concern is not one of too much soil N, but one of too much N in the wrong form (e.g. nitrate) and place (i.e. water supplies).

Annually, only about 3% of the total soil N is converted to a soluble, plant-available form (Bremner, 1965). For optimum crop production, this means supplemental N sources are often required. Such N sources include fertilizer, feedlot waste and manure, biological fixation, irrigation water, and wet-dry atmospheric deposition. The goal is to see that as much available N as prudently possible from the soil and supplemental sources is utilized by the crop, thereby minimizing N losses to runoff, leaching, denitrification, volatilization, deposition, etc. Considering that the current efficiency of the supplemental N sources is often < 50%, appreciable room remains for improvement. A prime mission of agricultural N researchers is to develop workable technology that will enhance this efficiency.

## N LOSS PROCESSES

## Wind Erosion

Legg and Meisinger (1982) estimated that 4.5 million metric tons of cropland N are lost annually to wind and water movement. Twenty percent, or 0.9 million tons, was attributed to wind. As with water, erosion by wind is a selective process, removing finer particles containing a disproportionately greater amount of N. Sparse data on N enrichment ratios for wind erosion exist, but would appear to be about 2 and 3 for organic and inorganic N, respectively (Hagen and Lyles, 1985; Zobeck and Fryrear, 1986). More exacting prediction techniques are under development (J. R. Williams, 1989, personal communication) and, until then, these values can be multiplied by the wind erosion amount to approximate N loss. The amount of wind erosion can be calculated by the equation (Woodruff and Siddoway, 1965):

$$WE = f(I, WC, WK, WL, VE)$$

where WE is the wind erosion amount, I is the soil erodibility index, WC is the climatic factor, WK is the soil-ridge roughness factor, WL is the field length along the prevailing wind direction, and VE is the quantity of small grain equivalent vegetative cover. The above equation has been modified by Cole et al. (1982) to calculate wind erosion on a daily basis.

## Runoff

Nitrogen moves in surface runoff both in the soluble and particulate forms (Sharpley et al., 1987). The dominant soluble forms are nitrate, which tends to move down the soil profile with the initial infiltrating water of a storm event, and ammonium, which tends to become attached to the soil's cation exchange complex. For many agricultural watersheds, then, soluble N concentrations in runoff are fairly low and well within potable limits (i.e. 10 and 0.5 mg L<sup>-1</sup> respectively, for nitrate-N and ammonium-N). In fact, the soluble N concentrations in runoff are often less than corresponding values in rainfall. This means watersheds may act as filters in removing soluble rainfall N from runoff, and suggests there would be little point in trying to

predict soluble N runoff losses on the basis of soil N characteristics. Nevertheless, there are cases when high soluble N concentrations may be observed in runoff. Instances are when a soil horizon barrier (e.g. fragipan) exists in the profile resulting in interflow (Lehman and Ahuja, 1985; Kissel et al., 1976) that reappears as surface runoff, and when a major runoff event occurs shortly after surface application of N fertilizer (Smith et al., 1988). Then, even high ammonium-N fertilizer concentrations may be observed (i.e.  $10^+ \text{ mg NL}^{-1}$ ). The situation is not long lasting, though, and often by the next runoff event soluble N runoff concentrations are close to background. In the Southern Plains, < 5% of the applied N fertilizer tends to be lost as agricultural runoff (Kissel et al., 1976; Smith et al., 1983). Similar observations have been made for other areas, including Iowa (Alberts et al., 1978) and Louisiana (Dunigan et al., 1976).

Particulate N (PN) losses in runoff occur primarily in the form of soil organic matter components associated with the suspended sediment. Such losses may be calculated using an N enrichment ratio (NER) approach as described by Sharpley et al. (1985) where:

$$\text{PN} = \text{Soil PN} \cdot \text{sediment concentration} \cdot \text{NER}$$

with soil units as  $\text{mg kg}^{-1}$  and sediment units as  $\text{g L}^{-1}$ . The NER is predicted by the equation (Menzel, 1980):

$$\ln(\text{NER}) = 2.00 - 0.20 \ln \text{soil loss}$$

where the units of soil loss for each runoff event are  $\text{kg ha}^{-1}$ . The above approach has been shown to provide realistic particulate N loss estimates for a wide range of agricultural soils on both an event and an annual basis (Smith et al., 1986). The annual particulate N losses ranged from almost nil to  $7 \text{ kg ha}^{-1}$ .

### Leaching

Nitrogen leaching in soils occurs primarily in the nitrate form. While some leaching of ammonium may occur in sandy soils, its leaching susceptibility is reduced by adsorption to the soil's cation exchange complex. Neither does leaching of soluble soil organic N components appear to be a major problem (Smith, 1987). Various approaches exist for estimating nitrate leaching in the field, and they range from the sophisticated to the simplified.

More sophisticated approaches, better suited for controlled research studies, generally comprise some form of the convection-dispersion equation (e.g. Rose et al., 1982):

$$\frac{\partial C}{\partial t} = \frac{D}{\partial Z^2} C - \frac{V}{\partial Z} C$$

where C is the concentration of nitrate in the soil water, D is the dispersion coefficient, t is the time variable, Z is the soil depth, and V is the average velocity of pore water. In some cases, a nitrate plant uptake factor, B, may also be incorporated (Smith et al., 1984). Provided the necessary input factors are available, the sophisticated approaches can describe nitrate leaching quite well. For the uncalibrated field situation, however, the input factors are not always available, and simplified, more practical nitrate leaching approaches are required.

Simplified nitrate leaching approaches utilize the center of mass "piston displacement" technique, which has a wide experimental and conceptual basis (Davidson et al., 1978; DeSmedt and Wierenga, 1978). The principal assumption is that soil water above field capacity initially present in the profile is displaced ahead of the water entering the soil surface. Incorporation of the piston approach with pertinent soil and climatic factors allows for estimating nitrate leaching depth potentials on a general basis. The following example illustrates one simplified predictive method (Smith and Cassel, 1989). Basically, it involves incorporation of a soil leaching potential with a climatic leaching potential.

The soil leaching potential reflects the water holding properties of the soil and its capacity to permit infiltration. The climatic leaching potential is illustrated in Figure 1. The data are for the Raleigh, NC area and represent mean monthly precipitation (P) and potential evapotranspiration (PET). Note that the mean monthly PET exceeds P from late April to mid-October. Consequently, this is a period when, on average, water would not be available for

leaching. On the other hand, P exceeds PET from November through March, thereby indicating good potential for leaching.

To estimate the probable depth of nitrate leaching for a particular location, only the soil water holding properties, the associated P and PET, and the relevant management factors are required. Then, a simple water balance format (Smith and Cassel, 1989) is constructed. Figure 2 shows the probable depth of nitrate leaching utilizing the format for nitrate fertilizer applied surface broadcast to a Norfolk sandy loam at corn planting time or the preceding fall. Obviously, the probable depth of leaching is much greater (approximately 180 cm) when the fertilizer was fall applied. Moreover, by spring planting time the fertilizer N is already at a depth below the expected corn rooting depth, and would have good potential for leaching to the ground water. With the spring application, however, the probable depth of leaching is < 30 cm even at harvest time. Provided excess spring fertilizer N has not been applied, little nitrate would remain for leaching at this depth.

In the above example, no allowance was made for preferential leaching down soil macropores, or nitrate exclusion in the subsoil (Smith, 1972). Both such phenomena may need to be accounted for with certain soils and conditions. However, the macropores can be particularly difficult to account for, because their formation and continuity are dependent upon various factors, such as earthworms, plant roots, and soil drying. Likewise, stony, fissured, and karst topography soils (i.e. sinkholes at or near the surface) may provide direct flow channels for rapid movement of water and associated nitrate. In the case of irrigation, it is simply added in as a precipitation contribution. If desired, any soil-derived nitrate contributions may also be factored in. Situations involving deep nitrate leaching in the vadose zone are considered by Pionke and Lowrance (1989) and Shaffer et al. (1989).

#### Tile Drainage

Millions of hectares of poorly drained U. S. soils have been converted to highly productive croplands by tile drainage. Letey et al. (1979) reviewed nitrate-N concentrations in tile effluents from 61 sites in six areas of California. Average site concentrations ranged from 1 to 196 mg L<sup>-1</sup>, with only about one-quarter of the sites averaging < 10 mg L<sup>-1</sup>. A recent review of N losses in tile effluents from the Corn Belt states of Iowa, Minnesota, and Ohio, has been conducted by Logan et al. (1980). They found annual nitrate-N losses were generally < 30 kg ha<sup>-1</sup>, but increased with N fertilizer applications above crop needs. Nitrate-N concentrations were above 10 mg L<sup>-1</sup> even when 20 kg fertilizer N ha<sup>-1</sup> or less were applied annually to continuous corn. On the other hand, nitrate losses and concentrations with alfalfa were very low. Therefore, periodic inclusion of alfalfa on the lands in continuous corn may be beneficial in maintaining nitrate losses in the tile effluents at more desirable levels. Little is known, however, regarding N losses when alfalfa is destroyed and the land is planted to other crops.

Because tile line effluent represents only a portion of the infiltrating water, the question may be raised regarding the fate of nitrate in that water bypassing the tile and continuing downward. Provided an organic energy source is present, the potential for denitrification exists, as observed by Gambrel et al. (1975) for a North Carolina coastal plain soil. Such process may not always occur, however, because Hallberg et al. (1986), observed in Iowa that some high, nitrate tile concentrations reflected water table concentrations and that of infiltrating water which recharged the aquifer.

#### Volatilization of Gaseous N

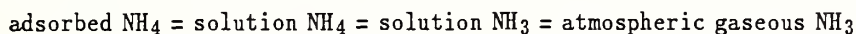
Gaseous N losses involve complex processes associated mainly with ammonia volatilization and denitrification. The former is more prevalent with surface applied N whereas the latter is more prevalent with incorporated N. Significant advances in gaseous N methodologies have been developed in the past 15 years. Techniques are now available for evaluating the magnitude of ammonia, dinitrogen, nitrous oxide, and nitric oxide losses as influenced by various cropping practices. Seldom, however, are all the gases measured at one time for any major cropping system



or cultural practice. A serious problem in making assessments is the spatial variability associated with soil heterogeneity. This brief review can not cover in detail the many gaseous N developments and their application. Additional information may be found in publications by Bock and Kissel (1988) Freney and associates (1983, 1988a, 1988b), Mosier and Heinemeyer (1985), and Terman (1979).

#### Ammonia

Freney et al. (1983) have reviewed the chemistry of ammonia and the soil factors that affect ammonia volatilization, retention, and transport. Ammonia is highly soluble, very reactive, and its volatilization increases with increasing pH. The various reactions which govern ammonia loss (Freney et al., 1983; Vlek et al., 1981) may be represented as:



Besides pH, equilibria among the various species are affected by temperature, partial pressure of  $\text{NH}_3$  in atmosphere, and  $\text{NH}_4$  concentration in solution. The effect of pH on the equilibrium between  $\text{NH}_4$  and  $\text{NH}_3$  can be calculated from the equation:

$$\text{pK}_a - \text{pH} = \log_{10} [\text{NH}_4]/[\text{NH}_3]$$

Emerson et al. (1975) provide an equation for calculating  $\text{pK}_a$  at a particular temperature:

$$\log_{10} K_a = -0.09018 - 2729.92/T,$$

where T is the absolute temperature ( $^{\circ}\text{K}$ ). The  $\text{pK}_a$  at 25 C is 9.246. Freney et al. (1983) indicated the relative percentages of  $\text{NH}_3$  at pH values of 6, 7, 8 and 9 are approximately 0.1, 1, 10 and 50 and, thus, as solution pH increases there is a greater potential for  $\text{NH}_3$  volatilization. Temperature also affects the solubility of ammonia in water and the diffusion rates in soil. As temperature increases diffusion rates increase, whereas ammonia solubility decreases. Its volatilization depends on the same environmental conditions as water evaporation. In fact, various investigators have shown that ammonia loss is directly related to water loss. Therefore, experimental set-ups to measure ammonia volatilization must not alter temperature, relative humidity, windspeed, etc.

Enclosures of various types have been used to measure ammonia loss in the laboratory and growth chamber with some success, however, field enclosures are considered unreliable because they interfere with windspeed, temperature, and water evaporation. It is known that ammonia is rapidly absorbed by plant leaves, so the size and development of the crop canopy influences atmospheric ammonia profiles. Also,  $\text{NH}_3$  is rapidly adsorbed to glass, rubber, plastic, and other surfaces. Hence, tubing and various materials used for enclosures are likely to retain the molecule. Many investigators have used open acid traps, acid saturated sponges, or filter papers, but such techniques interfere with air movement and create artificial diffusion gradients, making it nearly impossible to properly determine ammonia emissions.

At present, micrometeorological techniques that do not disturb the natural environment and integrate the ammonia flux over known areas are being used successfully to measure field emissions of ammonia from upland and flooded soils. Denmead (1983) describes the micrometeorological techniques of gradient diffusion, and mass balance. If there is a uniform source of  $\text{NH}_3$  at the ground surface over a large area, a concentration gradient is developed in the atmosphere above the area. And, if the conditions are constant with time, the upward transport of  $\text{NH}_3$  can be calculated from measurements of the gradient diffusion by the equation:

$$F = K \, dc/dz$$

where F is the flux density of  $\text{NH}_3$ , K is the diffusivity for  $\text{NH}_3$ , c is the atmosphere  $\text{NH}_3$

concentration, and  $z$  is the height above ground where  $\text{NH}_3$  is being measured. The equation is only valid when the flux is constant with height in the air layers above ground surface. This requires that the experimental area be large and uniform. The depth of the air layer in which the flux is constant with height is roughly between 1/100 to 1/200 of the fetch or distance upwind, thus limiting applications of the equation to fields of several hectares. The diffusivity,  $K$ , of  $\text{NH}_3$  in air is the eddy diffusivity (movement of parcels of air from one level to another) and is several orders of magnitude higher than molecular diffusivity. Also, its magnitude varies with environmental conditions and must, therefore, be measured in place. (Denmead, 1983).

Mass balance is probably the simplest and most convenient of the micrometeorological methods, and is suitable for small areas. It is used where the flux from a treated area is expected to be quite different from a similar size non-treated area. The horizontal flux of  $\text{NH}_3$  across a vertical plane of unit width on the downwind edge of a treated area is equated with the flux from a similar area upwind. The horizontal flux at any height is the product of horizontal wind speed and  $\text{NH}_3$  concentration. The product of wind speed and  $\text{NH}_3$  is modified by  $\text{NH}_3$  emission (Denmead, 1983). The flux density of  $\text{NH}_3$  can be expressed in the integral equation:

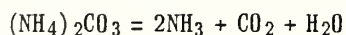
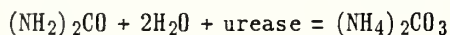
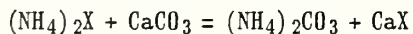
$$F = 1/x \int_0^z uc \, dz$$

where  $F$  is the flux density,  $x$  is the distance traveled by the wind over the treated area.,  $z$  is the height of the layer in the atmosphere,  $u$  is the wind speed, and  $c$  is the  $\text{NH}_3$  concentration in excess of non-treated or background. To evaluate the integral equation, the profiles of  $u$  and  $c$  must be precisely defined at a number of heights. However, if the wind has a log, or power law, profile it is possible to predict the flux from measurement of the  $\text{NH}_3$  at one height called ZINST. Denmead (1983) discusses ZINST in detail and the experimental evidence for its existence. Freney et al. (1985) have used simplified versions of the mass balance approach to measure ammonium volatilization from flooded rice at ZINST (0.8m), but the method has not been tested extensively for upland crops. They have also developed a bulk aerodynamic method to measure ammonia emission from flooded rice, but found the method was not as accurate, compared to mass balance.

As noted earlier,  $\text{NH}_3$  losses increase with increasing alkalinity or when ammonium salts produce microenvironments of high pH. In the dissociation of ammonium to ammonia:



there is a proton produced, so as ammonia is lost from the system the floodwater or soil media should become more acidic. The rate of acidification depends upon the buffering capacity (base saturation and calcium carbonate) content of the media (Avnimelech and Laher, 1977). Therefore, one can expect increased ammonia volatilization from calcareous soils that have a high base saturation, high carbonate content or other forms of alkalinity. There are a number of carbonate reactions that are important to ammonia volatilization (Terman, 1979; Fenn and Kissel, 1973; Fenn, et al. 1981; Stumpe, 1981):



Knowledge concerning the partial pressure of  $\text{NH}_3$  and  $\text{CO}_2$  that occurs upon the dissolution of ammonium carbonate or the hydrolysis of urea is necessary to predict or understand ammonia losses from flooded and upland soil. For example, the photosynthesis and metabolism of algae in floodwater can drastically alter the  $\text{CO}_2$  partial pressure with diurnal cycles occurring because of the cycling of solar radiation. Also, the anion ( $X$ ) associated with ammonium fertilizer influences the solubility of  $\text{CaX}$ . For example, if  $X$  is  $\text{SO}_4$  or  $\text{HPO}_4$  then  $\text{CaX}$  precipitates, driving the reaction toward ammonium carbonate, whereas, if  $X$  is  $\text{NO}_3$  or  $\text{Cl}$  the solubility of  $\text{CaX}$  is

increased and the formation of ammonium carbonate is reduced, thus decreasing the volatilization of ammonium.

Knowledge of the above information allows one to design a number of fertilizer practices that should reduce ammonia losses with ammonium salts, urea, and anhydrous ammonia. Ammonia volatilization can be slowed by restricting transport within the soil and evaporation at the soil surface. In both upland and flooded conditions research has shown ammonia losses can be reduced by placing ammonia based fertilizers below the soil surface or if the fertilizer is broadcast on the soil surface immediately incorporating it by cultivation. If ammonium salts or urea are to be applied in irrigation systems avoid times of high wind and high temperatures, i.e. apply water fertigation during periods of low water evaporation, in the evenings or at night when the temperatures and windspeeds are at their minimums. Apply enough water (at least 2.5 cm) to move urea down into the soil. On calcareous soils avoid ammonium salts, like ammonium sulfate, that react to form ammonium carbonate and insoluble precipitates with calcium. Each crop has a nitrogen requirement that varies as the crop matures. Applications of nitrogen timed closely to the crop requirement are most efficient. Usually, well established roots are more effective at reducing inorganic N sources in the soil profile. In addition, foliar absorption of ammonium in the crop canopy can further reduce ammonia losses (Denmead et al. 1976). For rice plants recovery of applied urea is known to be much greater if applied at maximum tillering or panicle initiation (Heenan and Bacon, 1987; De Datta, 1981).

#### Nitrogen oxides/Dinitrogen

Transfers of nitrogen oxides ( $\text{NO}$ ,  $\text{NO}_2$ ,  $\text{N}_2\text{O}$ ) from the plant-soil system have gained increasing attention in recent years. This is because  $\text{NO}$  and  $\text{NO}_2$  have been implicated in acid rain formation and the generation of photochemical smog, and  $\text{N}_2\text{O}$  has been implicated in the photochemical destruction of ozone in the stratosphere. All the nitrogen oxides plus dinitrogen ( $\text{N}_2$ ) are important nitrification and denitrification gaseous reaction products that decrease nitrogen use efficiency in crop production.

Various methods (Lemon, 1978; Denmead, 1979; Rolston et al., 1976; Ryden et al., 1978; Galbally and Roy, 1978; Galbally et al., 1987; Segal et al., 1982; Mulvaney, 1984; Mulvaney and Boast, 1986; and Blackmer et al., 1982) have been used for measuring emissions for nitric oxide ( $\text{NO}$ ), nitrogen dioxide ( $\text{NO}_2$ ), nitrous oxide ( $\text{N}_2\text{O}$ ), and dinitrogen ( $\text{N}_2$ ). Flux measurements using the meteorological techniques outlined above for ammonia haven't been very successful for nitrogen oxides and dinitrogen because analytical methods are not sufficiently sensitive to directly measure relatively small fluxes from the soil surface. The atmospheric reservoir of dinitrogen is extremely large and results in a tremendous dilution effect for  $^{28}\text{N}_2$  and  $^{29}\text{N}_2$ . The atmospheric reservoir for  $\text{N}_2\text{O}$  is approximately 300 ppb, and is still large enough to cause considerable dilution of small fluxes of  $\text{N}_2\text{O}$ . Nitric oxide is readily oxidized, so its lifetime in the atmosphere may be in the order of seconds to minutes.

To overcome the analytical difficulties, scientists have turned to chambers of various designs to enhance atmospheric concentrations (Denmead, 1979; Ryden et al., 1978). Hutchinson and Mosier (1981) vented an enclosed cover to the atmosphere so that atmospheric fluctuations are exerted on the soil, which may result in a pumping action on soil gases. As the nitrogen oxides or dinitrogen accumulate beneath the cover, the gas exchange between the soil and enclosed atmosphere should decrease. They modified the flux equation to overcome this problem by measuring the change in concentration of the gaseous component with time. The problem can be further improved by using short collection periods. However, short collection periods must then account for diurnal variations (Galbally et al., 1987; and Blackmer et al., 1982) that can be related to temperature changes in upland and flooded crops and the diurnal algal growth patterns, caused by diurnal light patterns in flooded rice. Furthermore, since fluxes are measured by enclosing very small areas of soil small errors associated with the measurements may be magnified as data are extrapolated to large land areas. Analysis variability is caused by such soil factors as carbon content, pH, temperature, nitrate content, water holding content, soil texture, and microbial ecology, which affect nitrification and denitrification. Also, the enclosure volumes

are typically quite small and can not accommodate large plants. Therefore, plant influences on loss patterns are usually not measured. These patterns may change as plants assimilate N, absorb water, and shade the soil. In the rice crop system the plant may be a major conduit for gaseous N transport through the flooded system. In addition little is known how enclosures exclude short-term pressure fluctuations associated with turbulent winds which might have effects on gaseous fluxes.

Nitric oxide and  $\text{NO}_2$  concentrations in an incoming air stream can be determined by chemiluminescent detection. Reactions are chemiluminescent when the excess energy contained in the products can be released in the form of light. The reaction between NO and  $\text{O}_3$  produces  $\text{NO}_2$  in an excited state, which relaxes to produce red light which can then be detected by a photomultiplier, permitting an NO determination in the order of 1 to 2 ppb. Nitrogen dioxide in the air stream can be detected by reducing it with a molybdenum reaction at 400 C to NO and then using a difference method to separate NO from  $\text{NO}_2$ . When nitrogen dioxide encounters a surface wetted with luminol (5-amino-2, 3-dihydro-1, 4-phthalazinedione) a chemiluminescence is produced, the luminescence coming from the luminol. In this case,  $\text{NO}_2$  can be measured directly at concentrations in the 10-20 pptv. Using the luminol method, NO must be oxidized to  $\text{NO}_2$  before detection and the separation of the two compounds is accomplished by difference.

From laboratory measurements it is clear that nitrification is a major contributor of NO. Hutchinson et al. (1987), in incubation studies, found that the addition of ammonia enhanced NO emission under aerobic but not under anaerobic conditions. The fraction of added ammonia lost as NO exceeded  $\text{N}_2\text{O}$  and was large enough to have important agronomic and environmental implications. Tortoso (1988) using inhibitors has shown that NO is produced during the oxidation of ammonium to nitrate and emission rates increased as nitrite concentrations increased. When present in the soil, nitrite can enter into enzymatic and nonenzymatic reactions, leading to NO,  $\text{NO}_2$ ,  $\text{N}_2\text{O}$ , and  $\text{N}_2$  production (Cady and Bartholomew, 1960; Clark, 1962; Clark and Beard, 1960; Clark et al. 1960; Chalk and Smith, 1983; Wijler and Delwiche, 1954; Nommik, 1956; Smith and Clark, 1960; Stevenson and Swaby 1964; Stevenson, et al. 1970; Nelson and Bremner, 1969; Porter, 1969). Blackmer and Cerrato, (1986) treated 28 autoclaved soils with nitrite and found amounts of NO ranging from 1 to 35  $\text{mg kg}^{-1}$ , statistical analyses indicating that 95% of the variability for NO production could be explained by considering only soil organic matter content, pH, and their interactions.

During the past decade there have been few field measurements of NO and  $\text{NO}_2$  made and the limited measurements make it difficult to judge their importance in the N cycle. Galbally et al. (1987) studied the emission of NO from fertilized rice and found only 0.002% of the fertilizer N was lost during the 10 day study period. They observed that the  $\text{NO}_x$  flux was very low for the first few days after application of urea and about 5 times higher during the period when significant nitrite and nitrate were present in the floodwater. Tortoso et al. (1986) found under well aerated conditions in non-fertilized systems that NO production followed the order of coniferous forest > native shortgrass prairie > fallow agricultural fields. They also noted that natural rather than agricultural ecosystems have a greater potential for aerobic loss of N oxides. They observed that NO emission rather than  $\text{N}_2\text{O}$  is the principal gas lost during nitrification and that the loss of NO during microbial denitrification may be higher than commonly believed. Hutchinson and Follett (1986) studied the effects of long-term cultivation practices on NO emissions and found the emission rates followed the order of native sod > no-till > stubble mulch > conventional plow treatments. Again, NO emissions were greater than  $\text{N}_2\text{O}$  emissions and were closely related to nitrification rates. Anderson and Poth (1987) found that disturbed ecosystems (burned and unburned chaparral) which display high rates of mineralization and nitrification exhibit losses of NO and  $\text{N}_2\text{O}$ . Blockage of nitrification by acetylene again demonstrated that emissions of NO and  $\text{N}_2\text{O}$  result from nitrification.

Nitric oxide and  $\text{NO}_2$  can be formed from nitrification reactions, chemodenitrification or self-decomposition of nitrous acid and denitrification (Smith and Chalk, 1980; and Nelson and Bremner 1970). The oxidation of NO and  $\text{NO}_2$  and hydration of  $\text{NO}_2$  during the diffusion of these gases through the soil results in some nitrate formation (Nelson and Bremner, 1970). Field measurement flux rates for NO and  $\text{NO}_2$  (Slemr and Seiler, 1984; and Delany et al., 1986) were found to be strongly dependent on soil surface temperatures with minimums in the morning and maximums

in the early afternoon. These investigators have shown that the soil can be both a surface and a sink for these gaseous N oxides.

Gas chromatographic analysis with an electron capture detector (ECD) or an ultrasonic detector, a carrier gas of 95% argon—5% methane and Q porapak columns is a sensitive and precise method for the determination of  $N_2O$  (Rasmussen, et al. 1976; Blackmer and Bremner, 1977). Mosier and Mack (1980) used 10 port switching valves and two porapak columns so only  $N_2O$  is passed through the ECD, thus preventing oxygen interfaces with the detector. They also back flush their column to remove freon, which after a few samples begins to leak from the column and causes background drifting. Gas samples containing 400 ppb  $N_2O$  can be determined with a precision of  $\pm$  3 ppb in 4 to 5 minutes. Space limitations do not allow us to discuss the other numerous methods that have been used to estimate  $N_2O$  fluxes, so the reader is referred to Ryden and Rolston (1983) and Mosier and Heinemeyer (1985), who describe the methods and their advantages and disadvantages. The closed cover method is used to obtain flux rates of  $N_2O$ , but suffers from the shortcomings mentioned above for  $NO$ .

Laboratory studies have provided extensive information on factors that affect evolution of  $N_2O$ , and a number of these factors have been enumerated previously. It is probably sufficient to indicate that there are three major sources of nitrous oxide. They are: (1) the interactions of nitrite with soil organic matter and pH (chemodenitrification); (2) the production of nitrous oxide during the oxidation or nitrification of ammonium under aerobic conditions; and (3) the production of  $N_2O$  as nitrate is biologically reduced under conditions  $>$  60% soil water holding capacity, low oxygen concentration, and a soluble carbon source to provide energy for the reaction. Since the emission of nitrous oxide may be derived from three totally different reactions it is difficult or essentially impossible to extrapolate laboratory measurements to field conditions where these three pathways may operate intermittently under variable soil conditions and substrate concentrations. Plants may further complicate the picture as they affect the pools of available nitrate and ammonium, alter the soil moisture content, excrete soluble carbon compounds and affect the oxygen status of the soil. For example, it is known in flooded rice fields that the hollow stem of the rice plant is a conduit for oxygen movement to the roots and  $N_2O$  and  $N_2$  from the roots to the atmosphere. The rhizosphere zone around rice roots may be a mosaic of oxidized and reduced areas. In the field, when soils are warm and mineralization and fertilization provide ammonium substrate, then  $NO$  and  $N_2O$  production proceed at a low steady rate as long as conditions are favorable to nitrification. This pattern may abruptly change to one of denitrification in a few hours following an irrigation or rainfall. Sharp fluxes from the soil surface may be observed within 24 to 48 hours but the peak rate depends on the ease at which various gases are able to diffuse to the surface and then there is a slow decline over days as aerobic conditions begin to return. Rapid uptake of nitrates by plants can limit the substrate for denitrification. Banded urea, anhydrous ammonia, and ammonium sulfate or phosphate can create zones where pH varies widely. In alkaline and poorly buffered soils the pH may increase drastically with increasing concentrations of these compounds. Nitrite oxidizers (*Nitrobacter*) growth is suppressed at higher pH, apparently due to ammonia toxicity, while ammonium oxidizers (*Nitrosomonas*) growth is not affected. This results in accumulations of nitrite with its possible chemodenitrification reactions (Chalk and Smith, 1983).

Nitrogen use efficiency and denitrification studies require direct infield estimates of dinitrogen and  $NO_x$ . Total gaseous emissions can be made with the aid of highly enriched N—15 salts or urea. The idea for the technique was first proposed by Hauck et al. (1958) and further expanded by Hauck and Bouldin (1961). The method involves applying highly enriched 20 to 99 atom % N—15 salts and urea to soil. If nitrification and denitrification products from the fertilizer give rise to gaseous N it will be selectively enriched, since the biological or chemodenitrification products are not randomly mixed with atmospheric dinitrogen or with the organic N of the soil. Using this non—random N—15 distribution method permits calculations (Siegel, et al. 1982; Mulvaney and Kurtz, 1984; and Mulvaney, 1984) of the N—15 enriched gas from fertilizer as well as gas from non—labeled soil N constituents. The gaseous products may all be reduced to dinitrogen, or they may be selectively separated by freezing techniques and measured separately.

Field measurements have shown large variability associated with  $\text{NO}$ ,  $\text{N}_2\text{O}$ , and  $\text{N}_2$ . There are seasonal patterns associated with rainfall or irrigation, plant growth variations, as well as substrate production from organic matter mineralization and these are compounded by fertilizer application timing and placement. As yet, there is considerable uncertainty about the seasonal or yearly emissions of dinitrogen and nitrogen oxides. However, information compiled by Gilliam et al. (1985) would indicate annual cropland losses for the two major gases,  $\text{N}_2$  and  $\text{N}_2\text{O}$ , are in the approximate order of 15 and 2 to 10 kg N ha<sup>-1</sup>, respectively. For  $\text{N}_2/\text{N}_2\text{O}$  evolution during denitrification, a ratio of 16: 1 has been used on occasion. In the case of  $\text{N}_2\text{O}$ , it should be noted that some may be lost from soil dissolved in drainage water, perhaps in amounts comparable to that lost in the gas phase at the soil surface (Dowdell et al., 1979).

## PRACTICES TO AMELIORATE N LOSSES OR INCREASE N USE EFFICIENCY

### Crop N Requirements

Crop N uptake determinations provide the basis for estimating N requirements which can then be adjusted to compensate for other N sources. The crop N requirement factor can be defined as crop N uptake near maximum yield (Meisinger, 1984). On a kg N uptake per 1000 kg grain basis, typical values for corn and wheat are near 25 and 40, respectively. The values vary somewhat by region, and may require site specific modifications to obtain economically and environmentally acceptable yields. Yield goals, when possible, should be based on prior history of the soil. A realistic yield goal is to use the average production over a 5-year period, and add no more than 5% to that production (Wiese et al., 1987). Additional details regarding impacts of cultural practices, methods of fertilizer application, crop rotations, and other considerations on crop development and N use efficiency have been compiled by Hauck (1984). Technological advances and more sophisticated management practices in the future will probably be required to address anticipated environmental concerns.

Efforts to improve N efficiency will require better synchronization between soil N availability and crop N requirements. Therefore, cultural practices, soil water status, crop pests, and many other factors that affect crop N uptake patterns will complicate management decisions. Genetic selection for improved N efficiency in crops such as corn and sorghum may reduce N requirements, but efforts to quantify N x genotype interactions can easily be dominated by environmental interactions (Duvick and Cavalieri, 1987). Efforts to modify plant N metabolic processes by manipulating genetic material in a way to improve N efficiency may be several decades away. Until then, computer simulation of crop growth, N uptake, soil biological and chemical processes, and water status provides an alternative way to evaluate management practices that impact nitrate leaching and ground water quality (McIsaac et al., 1984).

### Soil N Availability Tests

Available soil N represents residual nitrate in the profile, plus N mineralized from the soil organic matter during the growing season. While residual nitrate has proven to be a useful index in dryland regions (i.e. Plains states and West), no generally accepted index exists for N mineralization. Obviously, such development would represent a major advance for avoidance of excessive fertilizer N application.

A complement to a soil N test may be a plant tissue N test. An attractive feature of tissue tests (Rauschkolb et al., 1984) is that the plant root system tends to integrate spatial variability of soil N supplying power over a relatively large field volume. To date, tissue tests have found application mainly as a diagnostic tool to indicate N sufficiency/deficiency for high value, specialty crops, rather than field crops. Nevertheless, N tissue tests for cotton (Tucker and Murdock, 1984) and corn (McClenahan and Killorn, 1988; Schepers et al., 1988) appear to be receiving increasing acceptance.

### Residual Nitrate

Tests for residual nitrate are useful for soils suspected of having an appreciable accumulation of nitrate in the profile. These tests measure only that quantity of nitrate present at the time of sampling. Consequently, they have limited utility as a soil N index under conditions conducive to leaching. From a soil test standpoint (Whitney, 1970), profile nitrate should be sampled in row crop fields generally between November 1 and May 1. Winter small grains can be sampled after August 1 for preplant N application and after November 1 for top dressed N. Spring small grain fields would be sampled after October 1 until freeze-up, or in spring after the soil thaws. Additional considerations include sampling after pre-irrigation, and sampling sandy soils in the spring for row crops and top dressing of wheat. In all cases, the subsoil as well as the surface soil is sampled. Some laboratories will not conduct the nitrate soil test unless a minimum depth of 60 cm has been sampled. Also, depending upon the laboratory, more detailed information about the sampled area may be requested.

In the case of side dressing only, surface soil samples (30 cm depth) collected after corn planting (V6 growth stage) are being evaluated as a means to determine N application amount (Magdoff et al. 1984). Also, soil nitrate determination and N fertilizer application on-the-go have been reported for corn production in Illinois (Anonymous, 1989). An electronic sensor mounted on a moving soil shank measures nitrate directly and transmits a signal to a micro computer that controls the fertilizer N application.

#### Soil Organic N Availability

A significant portion of a crop's N needs is supplied by mineralization of soil organic matter during the growing season. An elusive goal has been to conveniently determine this amount of potentially available soil organic N, and use it as a base for a more precise fertilizer N recommendation. Various N availability indexes exist (Keeney, 1982), but they typically provide a qualitative rather than a quantitative measure of soil organic N availability. In the early seventies, Stanford and Smith (1972) approached the problem using a soil N mineralization potential ( $N_0$ ) concept. Their intent was to determine a soil's  $N_0$  under optimum conditions, and then adjust it for field moisture and temperature. In this manner, they hoped to be able to estimate how much soil organic N would become available during a prescribed growing season. Amounts of N mineralized for 39 agriculturally important U. S. soils were determined using a successive aerobic incubation procedure for 30 weeks. The  $N_0$  value for each soil was then estimated on the assumption that N mineralization obeyed first order kinetics, and the accepted value of  $N_0$  was that which resulted in the best fit for the linear relation between  $\log(N_0 - N_t)$  vs time. For surface soils,  $N_0$  ranged from 20 to over 300 mg kg<sup>-1</sup>.

Subsequently, considerable attention was devoted to determining short-term mineralization procedures for  $N_0$ , chemical extraction procedures for  $N_0$ , utilizing the concept to model N cycling in soils, and testing its application. Overall, application tests have been mixed, but with some success in situ (Herlihy, 1979), the greenhouse (Stanford et al., 1973), and the field (Oyanedel and Rodriguez, 1977; Smith et al., 1977). More recent tests indicate use of undisturbed soil in the mineralization procedure provides an  $N_0$  better suited to field conditions (Cabrera and Kissel, 1988). Also, a rapid steam distillation, borate buffer method of assessing potentially available organic N has now been introduced (Gianello and Bremner, 1988) that is not significantly affected by drying of the soil.

The original concept of  $N_0$  has also been modified by some workers (e.g. Molina et al., 1980) to include more than one soil organic N pool. However, recent work in Canada (Campbell et al., 1988) indicates a one-component  $N_0$  model is satisfactory for quantifying N fertilizer requirements. At present, also, attention is being given to an electro ultra filtration (EUF) approach to estimate  $N_0$  (Schepers and Saint Fort, 1988). Nevertheless, to date, no soil organic N availability procedure has received general acceptance from a soil test standpoint. Ultimately, a systems type, mass balance N approach (Meisinger, 1984) may prove to be the best alternative. Present recommendation is to follow pertinent state N fertilizer guides, which are geared to specific crop needs and soil areas.

## Sampling Variability

The importance of obtaining a representative sample for soil N characterization cannot be over emphasized. State recommendations for soil test purposes vary, but generally range from 10 to 20 composite surface samples per soil area sampled. For deeper profile samples, a fewer number may suffice because soil N variation and content tend to decrease with increasing profile depth. An aerial photo of the field in question is a good aid in delineating uniform areas for sampling purposes. These photos, available through SCS or ASCS offices on a scale of 20 cm per 1.6 km (8" per mile), have been taken during the dormant season. They often provide a clear delineation of the soil series, slope, drainage pattern, and land use. Such information, particularly when incorporated with Geographic Information Systems (Peterson, 1989), can be of considerable value in locating uniform field areas for sampling purposes.

Even small, relatively uniform soil areas can be quite variable, with nitrate coefficients of variation in the range 30—50% not uncommon (Keeney, 1986a). More sophisticated, geostatistical techniques for ascertaining the number of soil samples required for a given precision and accuracy may utilize the samples' spatial dependence (Nielsen et al., 1982). Such techniques involve the use of autocorrelograms and the concept of a regionalized variable. With the geostatistical approach, an estimate for the N characteristic at an unsampled site is determined from measurements made near by, rather than utilizing a site average. Application of geostatistics to soil science is still relatively new, and the results to date have often given answers similar to more conventional techniques (Warrick et al., 1986). A major drawback has been the cost, due to the greater number of samples required in the analysis.

## Nitrogen Fertilizers

A wide variety of N fertilizer formulations is available to producers to accommodate various times, rates, and methods of application. Anhydrous ammonia (gaseous) accounts for approximately 40% of all fertilizer N marketed in the U. S. (Hargett et al., 1988). This value increases to over 60% in Kansas, Nebraska, Illinois, and Iowa, largely because anhydrous ammonia is the least costly form of fertilizer N. Granular N fertilizers such as urea and ammonium nitrate, are easier to store and handle, but more costly. Liquid N fertilizers (e.g. urea ammonium nitrate, starter blends) are frequently the most expensive but more versatile in terms of blending with other fertilizers or herbicides, plus they can be easily injected into sprinkler irrigation systems. Fertilizer N formulations and methods of application are discussed by Tisdale et al. (1985), however, special fertilizer N management considerations may be required to minimize contamination of ground water. Management systems which hold promise for the future include the use of satellite imagery (e.g. Global Positioning System) and grid farming, which allow applying N inputs by soil variations rather than by fields (Reichenberger, 1989; Reichenberger and Russnogle, 1989).

The foremost consideration when developing N management practices to minimize nitrate leaching must be to determine when water is likely to percolate below the root zone. Irrigation is frequently implicated as the cause of nitrate leaching, however precipitation between April 1 and June 15 in much of the Northern Plains exceeds ET (Schepers, 1988). Therefore, the benefits of fertilizer N applications before or during this vulnerable period for percolation must be evaluated in terms of crop N requirements and risk of leaching. Nitrate leaching can be reduced when a nitrification inhibitor (i.e. nitrapyrin, DCD) is added to ammonium fertilizers to slow the rate of biological conversion to nitrate (Keeney, 1986b; Owens, 1987). Effects of nitrapyrin on corn yield are frequently positive when nitrate leaching losses would otherwise be large. In contrast, if early season leaching losses are small and the crop was not N deficient because of adequate or excess N availability, then crop response to nitrification inhibitors may be minimal (Hergert and Wiese, 1978; Touchton and Boswell, 1978; Nelson and Huber, 1978; Papendick and Engibous, 1978; Onken, 1978).

Starter fertilizers frequently promote enhanced early season growth, but may not affect yield because amounts of N in starter fertilizers placed near the seed are only a small part of the



total crop N requirement. Crop utilization is usually the greatest with sidedress fertilizers because application time and crop uptake are closely synchronized. Although sidedress N application may be the preferred method of fertilization, split applications or starter fertilizer may be required to meet early season crop N requirements. Somewhat greater broadcast N application rates are frequently required when crop residues have the potential to immobilize N as under no tillage conditions (Doran and Smith, 1987). For this reason, greater fertilizer N uptake by crops can be achieved with surface band applications or with subsurface knife applications. Crop clearance by tractors usually restricts fertilizer applications requiring large amounts of power, however, high clearance vehicles make it possible to apply some forms of fertilizer virtually anytime. Wheel-type spoke injector mechanisms mounted on elevated tractors or similar vehicles permit subsurface placement of liquid fertilizer at virtually any stage of crop growth (Baker et al., 1985). Foliar application of N fertilizer to row crops and small grains has had limited effects on yield. Below et al. (1984a, 1984b) found that tagged-N foliar fertilizer applied to corn at anthesis was recovered in the grain, but yield and N concentration were not affected by the foliar N application. In this case, the crop was not N deficient and the foliar absorbed N-15 substituted for other N in the crop.

Nitrogen fertilizer, along with genetic improvement and irrigation, are probably the three most significant factors responsible for yield increases over the past 5 decades (Carlone and Russell, 1987). Cultivar selection for crops requiring large amounts of N, such as corn, has traditionally been made under high fertility conditions. Occurrence of nitrate in ground water has prompted interest in more N efficient cultivars. Corn yields from most N x genotype studies are not conclusive, largely because environmental interactions are difficult to quantify and frequently dictate the nature of the yield response (Duvick and Cavalieri, 1987). Therefore, research to select genetically superior cultivars for improved N efficiency is proceeding cautiously as the potential for improvements using genetic engineering and molecular biology techniques are evaluated. Both field (Olsen, 1986) and hydroponic studies (Below and Gentry, 1987) indicate the benefit of mixed ammonium and nitrate nutrition for corn. Metabolic reasons for the increased corn yield attributed to mixed N nutrition are uncertain (Hageman, 1978), but cultivars respond by increasing kernel number, others by increasing kernel size, and some a combination of the two (Schepers and Below, 1988). Nitrification inhibitors can be used to modify the proportion of nitrate to ammonium in soil on a temporary basis, but the proportion of these N forms may be adequate under natural conditions, depending on soil chemical and physical conditions.

Time of fertilizer application can have a major effect on yield and an implied effect on nitrate leaching. Best results for rain fed winter wheat in Colorado were obtained by applying N early enough to encourage leaching so that a majority of the stored water and N were available at similar depths in the root zone (Smika and Grabouski, 1976). Under irrigation, greatest N utilization by corn is usually obtained with sidedress application, which corresponds to the time of greatest N uptake. Fertigation can also be timed to coincide with the time of greatest crop N requirement, but may result in volatilization of ammonia sources (Denmead et al., 1982) and the convenience of fertigation may promote N application beyond the time of greatest utilization. As noted earlier, crop N status during the growing season can be evaluated by tissue testing procedures. Such procedures are routinely used for some fruit and nut crops, sugar beets, and potatoes. Application of tissue testing procedures to improve N management holds promise for other crops such as corn and sorghum (Schepers and Below, 1988) because leaf analysis for total N is an easy and rapid way to monitor crop N status, which is a reflection of N availability, regardless of the source.

Mineralized N is the most difficult source of N to quantify because of its dependence on tillage, climate, and substrate availability. Nevertheless, mineralized N can amount to a large part of crop N requirements on fertile soils, and if not utilized can contribute to nitrate contamination of ground water. The rather slow and perpetual nature of mineralization and immobilization requires special consideration so that it can be synchronized with crop N requirements when large amounts of crop residue are added to soil. Residues with < 1.2% N usually immobilize N (Broadbent, 1978) which reduces the potential for nitrate leaching, but it

can also create a N deficiency before mineralization dominates. Green manure cover crops, legume vegetation and animal manures usually contain adequate N to promote mineralization shortly after soil incorporation. The fertilizer N equivalence for legumes ranges from  $< 20 \text{ kg N ha}^{-1}$  for some interseeded legumes (Hesterman, 1988) to over  $100 \text{ kg N ha}^{-1}$  for several winter cover crops (Frye et al. 1988). Additional information regarding N use and crop residues may be found in Power and Doran (1984).

### Nitrogen/Soil Conservation Practices

#### Conservation Tillage

Conservation or reduced-tillage continues to increase in usage as an alternative for practically all forms of crop production (Follett et al. 1987). Management systems which maintain crop residues at or near the soil surface have several attractive features, including less on-farm energy use, more available soil water, and reduced soil erosion. Concurrently, the adoption of conservation tillage practices has stimulated interest in associated N environmental impacts.

There is no question that conservation tillage is effective in reducing particulate N losses associated with runoff (Gilliam and Hoyt, 1987). Berg et al. (1988), for example, on highly water-erodible wheat soils in the Southern Plains, observed a 21- and 8- fold reduction in sediment and total N loss, respectively, on no-till compared to conventional-till (disking and sweeps). This was over a 2-year, above average rainfall period. The N loss reduction was not as great as the sediment loss reduction due to humus enrichment, selective removal of fines, and less dilution by infertile subsoil. In the case of soluble N in the runoff, differences between the tillage treatments were not that great,  $1.7$  and  $1.9 \text{ kg NO}_3\text{-N ha}^{-1} \text{ yr}^{-1}$ , for conventional and no-till respectively. Such differences are generally small, provided the fertilizer N is incorporated in the soil (Gilliam and Hoyt, 1987).

Effects of conservation tillage on leachable N are not so well delineated as for surface runoff N. Generally, conservation tillage provides a wetter, cooler, more acidic, less oxidative soil environment. Under such conditions, the processes of ammonification and denitrification may be favored over nitrification. On the other hand, for that nitrate present, the leaching potential may be greater under reduced tillage. This is because more undisturbed, large soil pores exist for nitrate movement. Dick et al. (1986), for instance, observed twice as much water flow down, out of the root zone in no-till compared to conventional-tilled soils. This higher flow was attributed to reduced evaporation from the soil because of the surface residues and increased number of channels continuous to the soil surface. The surface mulch enhanced the environment for earthworms and the lack of tillage preserved for several years the flow transmitting capacity of their burrows.

The role of macropores regarding leaching under conservation tillage can be illustrated by two contrasting examples. In the first case, nitrate leaching was found to be less under no-till as compared to conventional-till, when nitrate was distributed on the soil surface (Kanwar et al., 1985). Under no-till, much of the water flowed through macropores to the subsoil, leaving most of the remaining nitrate distributed throughout the soil surface. Under conventional-till, no large macropores or channels leading from the soil surface to the subsoil were present, so the infiltrating water moved down the soil profile as a front, carrying the nitrate with it. The net result was less nitrate leached under no-till. In the second case, nitrate leached deeper in a killed sod, no-till system than under conventional-till (Tyler and Thomas, 1977). In this case, soluble ammonium nitrate fertilizer had been applied to the soil surface. Rainfall dissolved the fertilizer, and the water flowed through root channels from the soil surface, carrying the nitrate deeper down the profile. For the two examples, then, we see that the nitrate leaching process can be very different for similar tillage systems, depending on whether the fertilizer finds its way to the macropores.

Equipment development and research are currently under way to precisely and accurately position N fertilizer under conservation tillage to reduce soluble N leaching (Baker, 1985). For

example, in the case of ridge--till there may be an advantage in placing the N fertilizer in the ridge, because a large part of the water infiltrates in the valleys between the ridges. Another possibility may be in placing the fertilizer under the soil surface, in a coarse-textured band (Bowers et al., 1975).

#### Rotations, Cover, and N Scavenging Crops

Rotations and cover crops, historically used as a means of conserving soil and/or providing an organic N source, have received renewed interest as an aid in avoiding excessive N losses to the environment. Whereas monocultures of grain crops (e.g. corn and wheat) require high inputs of fertilizer N, such inputs can be reduced with crop rotations which require less or produce organic N (Papendick et al., 1987). Voss and Shrader (1984) note that first year, non N fertilized corn in Iowa following alfalfa may outyield fertilized monoculture corn. Concurrently, because less excess profile N may be expected with a rotation, there should be less potential for N leaching. An exception may be under certain rotation--fallow conditions designed to conserve water in drier areas. The saline seeps of the Northern Plains, for example, are a result of a rotation--fallow practice allowing deeper water penetration that carries native solutes, including any profile nitrate present, to a shallow impermeable layer. The solutes may then move to a downslope position, resulting in the seep (Olson, 1986). It should be noted that such seeps are now being observed with increasing frequency in the Southern Plains (Naney et al., 1986), especially on terraced fields.

Winter cover crops can be effective in absorbing both nitrate and available water during the fall, winter, and spring, thereby reducing the N leaching potential. When the cover crop is returned to the soil, some of the absorbed N is then available to the following crop. Both legumes (e.g. vetches, clovers, peas) and non legumes (e.g. small grains) are used, but from a strictly N leaching standpoint, rye would appear to have certain advantages. It is the most cold tolerant, with lodging and disease posing little concern.

While an annual crop, such as rye, can be effective in scavenging excess available N from within crop rooting zones, deep-rooted perennials should be considered for nitrate accumulations below normal rooting depths. Alfalfa, with a potential rooting depth in excess of 5 meters, is a crop which merits particular attention (Olsen et al., 1970; Schertz and Miller, 1972; Stewart et al., 1968). In fact, soil nitrate contents under alfalfa can be lower than under native grasses (Table 1).

#### Filter Strips

Vegetative filter strips, also referred to as buffer strips and riparian zones, are defined as strips or areas of vegetation for removing sediment, organic matter, and other pollutants from runoff and waste waters (SCS, 1984). Under field conditions, excess runoff from terraces is frequently diverted to a strip. Upon entering the strip, both the flow velocity and transport capacity of the runoff are reduced. The sediment and its associated pollutants are then removed from the runoff by filtration, deposition, infiltration, sorption, decomposition, and volatilization processes. The size of a filter strip needed for a particular situation may be calculated (Ohlander, 1976), but 30 m seems to be a common width.

The effectiveness of filter strips in removing sediment and particulate waste N is well established (Neibling and Alberts, 1979; Lowrance et al., 1984; Robinson and Brockway, 1980; Young et al., 1980). For example, Robinson and Brockway (1980) evaluated filter strips on 18 research plots and observed that a 2.4 m small grain strip achieved sediment removal rates of 80+% from furrow irrigated beans, and reduced the mean particle diameter in the runoff sediment from 64 to 17  $\mu$ m.

Results regarding the effectiveness of filter strips for removing soluble N forms in runoff are mixed. Omernick et al. (1981) question such effectiveness and point out soluble nutrients associated with sediment may move to streams over time by subsurface flow. Lowrance et al. (1983, 1984) studied water borne nutrient budgets for the riparian zone in an agricultural area. They

found the riparian zone acted as a filter for nitrate, which was transformed to organic form. Similar results have been reported in New Zealand (Howard—Williams et al., 1986) where more than 70% of the nitrate uptake over time was attributed to aquatic macrophytes in the riparian zone. Obviously, some denitrification may also be occurring. Another factor influencing the efficiency of a filter strip in removing soluble N forms is how fast they pass through the vegetation. Incorporation of an N scavenging crop, such as alfalfa, in the filter strip may retard vertical movement of the soluble N.

#### Terraces, Settling Basins and Impoundments

These structures also provide a means for trapping particulate and soluble N forms in runoff. Schuman, et al. (1973), for example, observed that N losses from level—terraced corn were only one—tenth those from contour—planted corn. In the case of settling basins, Edwards et al. (1980, 1983) channeled runoff from a 243 m<sup>2</sup> beef feedlot through a settling basin and determined the basin reduced particulates in outflow by 54%. For major storm events, the basin was more effective than filter strips. Tile drainage of small sediment basins built into terraces also allows the discharge water to be relatively low in particulates and nutrients (Schepers et al., 1985).

With large impoundments, such as lakes and reservoirs, the emphasis is more on maintenance of a high quality water supply for potable, irrigation, and recreation purposes, rather than use as a settling basin. Nevertheless, even these type structures serve a major role in the settling and removal of N components. For example, the geometric mean of total N concentrations in waters of 87 European and North American lakes was determined to be about one—half that in inflows, with the reductions in concentrations generally being greater for longer residence times (OECD, 1982). Less information is available for smaller agricultural field impoundments, but Menzel et al. (1986) recently determined concentrations of nitrate, ammonium, and Kjeldahl N in runoff and two associated SCS impoundments. The impounded water and outflows had lower N concentrations, except for similar ammonium concentrations. Total N (Kjeldahl plus nitrate) concentrations in the outflows were about 20% of that in the inflows. Similarly, Olson et al. (1973) observed that nitrate N content of inflow to three Nebraska reservoirs averaged 1 mg L<sup>-1</sup> whereas the nitrate N content of outflow was only 0.05 mg L<sup>-1</sup>.

#### Irrigation

Approximately 17% of all cropland in the U. S. is irrigated (Olson, 1986). Irrigation is usually intended to supplement precipitation or to leach excess salts from the root zone. While irrigation may reduce the risk of crop failure, it can subsequently increase the potential for nitrate leaching and risk of ground water contamination. Unfortunately, soils most susceptible to leaching are often those that are irrigated. The most positive approach to controlling nitrate leaching is to optimize the leaching fraction. Where soil salinity is a factor, the leaching fraction should be just sufficient to maintain an appropriate salt balance. Both time and method of irrigation must be considered as an integral part of N management practices.

Irrigation methods that rely on gravity for distribution of water (e.g. furrow and border) typically result in temporal and spatial variability in application rates (Elliott and Walker, 1982), especially on nearly level fields where water spreading may be a problem. Soils with moderate to rapid infiltration rates accentuate water distribution problems and can lead to portions of the field that are under water stress while in other areas the crop can exhibit N deficiency symptoms because of nitrate leaching. Practices to improve water application uniformity of gravity systems include reducing the length of run, land grading to establish uniform slopes, and reducing surface roughness to increase the rate of water advance across the field (Kemper et al., 1982). Efforts to increase uniformity of water application with gravity irrigation systems are frequently coupled with runoff reuse systems. Recently, computer operated diversion valves in gated pipe distribution systems (surge irrigation) have made it possible to apply water at high rates for short periods of time to rapidly wet the soil and

thereby promote soil sealing (Miller et al., 1987). Subsequent water application can be timed to match the desired application rates with the soil water holding capacity of the root zone to minimize both percolation and runoff.

Runoff water from furrow irrigated land is usually only slightly enriched in nitrate as it passes across the field compared to nitrate concentrations in the water source. Similar trends are noted for nitrate enrichment of runoff originating from precipitation, while ammonium in any water source is usually sorbed as it passes across the landscape (Schepers and Fox, 1989). The exception, regardless of the water source, is runoff generated immediately after surface application of N fertilizer or manure (Schepers and Francis, 1982). Uncertainties regarding prediction of nutrient discharge in runoff arise because of differences in antecedent soil moisture conditions and the degree of mixing during the runoff process (Wallach et al., 1988).

Other methods of water application such as level basin and sprinkler irrigation are usually tailored to a specific terrain or type of crop. Sprinkler irrigation systems often provide the greatest flexibility in terms of time and rate of application, which should minimize the risk for nitrate leaching. Many high pressure sprinkler systems which are costly to operate have been replaced by medium and low pressure systems to reduce energy costs, however, these systems intuitively lead to additional runoff and/or infiltration (Gilley and Mielke, 1980). The relatively uniform water application possible with sprinkler systems makes them ideal for application of N fertilizer (fertigation) or pesticides at a time when the crop is too large for traditional application methods. Properly timed N applications in small amounts of frequently applied water can reduce leaching losses (Rhoads and Stanley, 1981) and should place less risk of contamination on ground water than preplant N applications. It is important to note that precipitation immediately following irrigation can result in excess soil water and subsequent leaching, regardless of how carefully irrigation is managed. For this reason, irrigation scheduling procedures frequently attempt to maintain a small storage reserve within the root zone to accommodate a limited amount of precipitation without causing leaching.

Drip or trickle irrigation is probably the most water and nutrient efficient method of irrigation, but it is also the most costly to install (Mitchell, 1981). Therefore, it is frequently used on high value crops (e.g. orchards, vineyards, strawberries) where installation cost can be amortized over several years and where quality of the saleable product can be reduced by sprinkler application and where water distribution and/or soil aeration is a problem with gravity irrigation. Nutrient additions to soil through drip irrigation systems should minimize nitrate leaching provided excess water applications are not required to maintain a leaching fraction to regulate salt accumulation.

Special water management considerations are required near the end of the growing season when precipitation during the fallow period is likely to cause leaching. Therefore, irrigation scheduling procedures should attempt to end the growing season with as large a water deficit as possible without reducing yield. Coarse textured soils are especially difficult to manage with respect to nutrients and water because of the frequently shallow root systems. For these reasons, rate and time of water application must be determined for each soil, method of irrigation and type of crop. Fertigation can be a valuable nutrient management tool, but usually requires additional technical information regarding the timing, form and amount of N addition to synchronize soil N availability with crop N requirements (McIssac et al., 1985).

Ambient levels of nitrate in irrigation water contribute to the inorganic N pool in soil, but fertilizer credit given for water borne nitrate may be overestimated if irrigation occurs late in the growing season after a majority of the required N has been assimilated by the crop. Tagged-N studies indicate this is the case for nitrate water applied to corn in July and August (Francis and Schepers, 1986; Varvel et al., 1985). Therefore, fertilizer or other N sources, which are subject to leaching by spring precipitation, are frequently required to meet early season crop N requirements. Late season irrigation N applications may be effectively utilized because N uptake by many field crops precedes dry matter accumulation. Although nitrate in irrigation water can be utilized by crops, its contribution to yield should be assessed relative to other N sources and synchronized with crop N requirements. Information about nitrate in the drainage waters associated with irrigation can be found in MacKenzie and Viets (1974).

## Well Protection

Many groundwater contamination problems with nitrate in rural areas simply involve direct surface runoff into older, hand-dug or newer, improperly installed, drilled wells (Smith et al., 1987). In such cases high ammonium contents may also be observed. Cloudy, organic matter containing well water is a good clue that surface contamination is involved. Then, bacterial determination (i.e. coliforms) may be in order. This is because infantile methemoglobinemia can be a complication of enteritis (bacterial origin), even when nitrate intake is negligible (Bryson and Boeckman, 1989).

With drilled wells, the problem is usually an improper seal, which allows surface runoff into the annulus between the borehole and casing. Groundwater seepage also tends to follow the same general direction as that of surface water. To the extent possible, agricultural field, feedlot, farmstead, and septic tank drainage should be directed away from the well. In the case of feedlots, most nitrate problems occur when they are not in use. Then the surface seal breaks down, conditions become more aerobic, and nitrate production from the accumulated ammonium is initiated. Also, to be avoided around wells are the storage, mixing, and loading of N fertilizer.

## N RESEARCH TOOLS

### Tracers

Nitrogen tracers provide the only definitive means for determining both the fate and behavior of applied N in the environment (L'Annunziata and Legg, 1984; Vose, 1980). A basic assumption is that N isotopes behave in an identical manner chemically, physically, and biologically. Also, natural N is assumed to have a fairly constant N-15 isotope composition. In the case of most N-15 enrichment field studies, these assumptions cause no problems. Currently, both N-15 enriched and depleted (i.e. essentially 100 atom percent N-14) fertilizer materials are available for field research.

The sensitivity of detection for stable N isotopes is relatively low due to analytical limitations and some natural variations in isotope abundance. To distinguish isotopically enriched from natural N, the N-15 abundance in the sample must lie outside the range of natural variations (approximately  $0.366 \pm 0.004\%$ ). Consequently, the permissible dilution of N-15 enriched samples is limited to 100/0.004 or 25,000 times its weight, and that of N-15 depleted samples is even less. Various instrumentation options are now available for isotope analyses. For less exacting work (1 to 3% precision), relatively inexpensive and simple N-15 emission spectrometers are available. For high precision work (< 0.2%) the isotope ratio mass spectrometers can be used. In addition, highly sensitive, automated commercial mass spectrometry analysis is now available, capable of analyzing 200+ samples per day.

Actual N-15 enrichments in fertilizer studies have varied from about 1 to nearly 100%, but are usually in the order of 5 to 10% for most routine fertilizer efficiency studies. For those field studies involving N cycling (e.g. mineralization-immobilization, leaching, gaseous N emission) and a complete N fertilizer balance, much higher enrichment may be used to provide for the enormous dilution effect. Depending on the length and conditions of the study, from 10 to nearly 100% enrichment may be desired.

Large quantities of N-15 depleted materials are now available as a by-product from N-15 enrichment for use in field scale studies. Practical considerations center around analytical sensitivity and isotope dilution. Generally, the use of N-15 depleted materials is confined to single-season, plant uptake studies, or those simply following the movement of applied N from soil to water.

Variations in natural N isotope abundance have been used in some N cycling and environmental studies, but the analytical and procedural problems are considerable. This is because, as noted above, the natural variations are extremely small and, in addition, may change during N transformations (Broadbent et al., 1980; Smith et al., 1983). To date, the approach has not received general acceptance.

The balance sheet approach with N-15 enrichment offers the most complete way for determining the disposition of applied N in the environment. The proper experimental method depends upon the resources available, the objectives, and the N transformations of interest. Due to costs, sampling problems, and spatial variability, N-15 field studies are typically confined to small plots, often < 1m<sup>2</sup>. Sampling and spatial variability can be critical, due to N transformations associated with water content and movement. Small plots allow complete removal of the soil, thereby avoiding problems due to coring and spatial variability. However, such removal does destroy the plot for future sampling. Small plots also tend to restrict fertilizer disposition studies to fibrous-rooted plants. Often, the plots are enclosed with a casing material to prevent lateral movement of the fertilizer, and may be instrumented with various probes and sampling devices. Indiscriminate use of such substances should be avoided, because they may influence the N transformations and thereby vitiate the results (Smith and Young, 1984). In addition, it should be noted that some soils are more suited to N lysimetry studies than are others. For example, special care should be exercised in the case of swelling clay soils, because enclosure may enhance both surface ponding and vertical movement down the lysimeter walls.

For those cases where N isotopes are too expensive to follow the potential path of soluble N (i.e. nitrate) movement in a field or watershed, a substitute tracer may be used. Bromide is the substitute tracer that has received the most attention. It moves very similarly to nitrate, has generally low natural occurrences, and is not regarded as particularly toxic to plants (Smith and Davis, 1974). Caution should be exercised, however, if livestock are confined to pastures receiving high rates of bromide (Owens, Van Keuren, and Edwards, 1985).

#### Models

Agricultural nitrogen models represent descriptions of how soluble, particulate, and gaseous N forms are transported under field conditions. They should not violate basic physical, chemical, or biological processes but by necessity, due to various knowledge gaps, include empirical relations. Often, the basic component involves a hydrologic model that has a submodel for N transformations. Comprehensive N models are complex, and field data necessary for adequate testing are not always available. Nevertheless, models can serve as useful basic and applied research tools to integrate multidisciplinary aspects, determine relative management change impacts, and pinpoint where specific information and understanding are lacking. In fact, limited process N models are now moving from the research to the actual field implementation stage.

One of the early ARS models that incorporated an N transport feature was ACTMO, introduced in 1975 by Frere et al., and later expanded upon by Stewart et al. (1976). Further development occurred in 1980 when CREAMS, probably the most widely used and successful general agricultural chemical, field-scale model, became available (Knisel, 1980). Since then, several other models with N transport and transformation features, in some cases extensions or modifications of those above, have come on the scene. Examples include AGNPS, DRASTIC, EPIC, GLEAMS, LEACHMN, NCSOIL, NITWAT, NLEAP, NTRM and RZWQM. Space limitations preclude a detailed consideration of the models here, so only a brief representation follows. For additional information on N models involving soil-plant-water relationships, reference is made to Frissel and van Veen (1981) and DeCoursey (1989).

Agricultural Non-Point-Source Pollution Model is a single-event, cell-based model developed to simulate sediment and nutrient transport from agricultural watersheds in Minnesota (Young et al., 1987). Basic components include hydrology, sediment, and N transport. The N component is subdivided into soluble and particulate forms. Output can be examined for either a single cell, or the whole watershed. At present, use of the model is being extended beyond Minnesota.

DRASTIC represents an empirical standardized system for evaluating ground water pollution potential utilizing hydrogeologic settings (Aller et al., 1987). The term is an acronym that stands for the 7 parameters the National Water Well Association considers to be the most important mappable factors controlling ground water pollution. They are D - depth to water, R - net recharge, A - aquifer material, S - soil, T - topography (slope), I - impact of the vadose zone, and C - conductivity of the aquifer. These factors are incorporated into a relative

ranking system that uses a combination of weights and ratings to produce a numerical index. Field testing of the approach has been lacking, but recent application to the Big Spring, Iowa, ground water basin was disappointing (Curry, 1987). Using nonparametric statistics, DRASTIC was applied to one set of nitrate data throughout the basin. There was no evidence of correlation with field data. Lack of correlation was attributed to failure of the weighting system or inappropriate methodology for the karst area involved.

Erosion Productivity Impact Calculator (Williams et al., 1984) was developed to determine the relation between soil erosion and soil productivity, and is capable of simulating long periods (e.g. > 50 years). EPIC is a comprehensive model that comprises nine major divisions: hydrology, weather, erosion, nutrients, plant growth, soil temperature, tillage, economics, and plant environment control. The N nutrient section includes surface runoff, leaching, denitrification, particulate N transport, mineralization, immobilization, crop uptake, fixation, and rainfall. The whole model is operational, but additional testing is underway for the N portion. Recent field tests with the nitrate leaching portion have been encouraging (Williams and Kissel, 1989). Tests involving N uptake by plants (Godwin and Jones, 1989) indicate N accuracy is highly dependent upon growth and yield components, and how well initial soil conditions are known.

Groundwater Loading Effects of Agricultural Management Systems (Leonard et al., 1987) was developed for field-size areas to evaluate the effects of agricultural management systems on the movement of agricultural chemicals within and through the plant root zone. The model utilizes the physically based CREAMS, and incorporates a component for vertical flux of the chemicals. Results of model testing demonstrate that GLEAMS simulates bromide movement and leaching within the general range of field data variability. Therefore, it should have utility also for simulating potential nitrate movement and leaching.

Leaching Estimates And Chemistry Model Nitrogen is a process — based model of water and N movement, transformations, plant uptake and N reactions in the unsaturated zone (Wagenet and Tson, 1987). Simulations have been formulated to describe the leaching of N fertilizer. They include volatilization of  $\text{NH}_4$ , and consider N uptake by plants to be a function of root density,  $\text{NH}_4$  and  $\text{NO}_3$  concentration.

Nitrogen and Water management (McIssac et al. 1985; Watts and Hanks, 1978) has been developed especially for corn on sandy soils. N transformations and transport are treated in relation to crop growth under given weather and irrigation conditions. The model results for plant N uptake, in the soil, and leaching of nitrate from the root zone were shown recently to compare favorably with field tests (Ahuja et al., 1989).

Nitrate Leaching and Economic Analysis Package (Shaffer et al., 1989) is a computer application package developed to obtain estimates of potential nitrate leaching from agricultural areas and projected impacts of this leaching on associated aquifers. Management practice, soil, climate, and economic impacts are required for application. Calibration and testing of the model has proceeded, utilizing results from sites in Nebraska and Michigan.

Nitrogen Tillage and Residue Management (Shaffer and Larson, 1987) is a comprehensive model with emphasis on management of nitrogen sources at the soil surface in conventional and reduced tillage systems. N transformations and transport are processes treated in detail, utilizing the NCSOIL submodel which contains both active and passive N pools. Analysis to date has been mainly with laboratory incubation data, but field testing is now underway.

Root Zone Water Quality Management (Rojas et al., 1988) is a comprehensive model that includes macropore flow and a description of the N cycle. It requires a large soil physical and chemical data base, has a semi user—friendly input format, and attempts to use an expert systems approach. Development is in progress, and the model is expected to have utility for comparing alternative management practices and their susceptibility for ground water contamination.

Accurately modeling the transformations and transport of N in the environment will remain a formidable and challenging task. This is because the behavior of N in the environment is so complex. Its valence can range from +5 to -3; it exists in both organic and inorganic forms; and it occurs in gaseous, liquid, and solid states. Moreover, it is highly influenced by temperature, water, microbial, and plant processes. Consequently, any N model will continue to contain a certain amount of empiricism and estimation, especially in the case of comprehensive N



models. Perhaps those that will prove to have the most field application are the ones that only rely on one or two N processes, such as leaching or volatility. As implementation of best agricultural N management practices proceeds, the need for applying comprehensive N pollution models may diminish. This is because there should be less N available for pollution. Consequently, the primary focus on N models may shift from an environmental to a fertilizer economics perspective.

#### RESEARCH NEEDS

During our preparation of this report various N research areas requiring additional attention surfaced, or became more obvious. Perhaps the most striking general need is that for an integrated, or holistic, field-based approach to assessing and managing N losses to the environment. Usually, field watershed studies have tended to consider only one specific avenue of N loss, such as leaching or runoff. Rarely are two or more avenues considered simultaneously, thereby making it difficult to assemble a comprehensive picture of N behavior. Consequently, a general recommendation is that representative watersheds be established and instrumented in certain major land resource areas where N fertilizer usage is concentrated. Instrumentation would involve devices for studying mechanisms and processes involved with N losses to surface runoff, groundwater, any tile drainage, and the atmosphere. The watersheds could be managed to reflect both rotation and monoculture cropping practices of the area, with a goal of maintaining acceptable yields and minimizing N leakage to the environment.

In the case of specific research needs, we bring into focus several below, some of which are being actively pursued by N researchers. No particular attempt has been made to place the needs in any type of national priority, because they vary somewhat according to area and management.

1. Assess and manage any deleterious N effects of conservation tillage on water quality and the atmosphere, under various soil, climatic, and management conditions.
2. Increase investigation of soil macropores' role in leaching of N. When necessary, devise N leaching submodels that take some quantitative account of macropore influence.
3. Increase N balance—field studies utilizing tagged N fertilizers, crop residues and/or animal wastes to assess specific avenues and extent of N leakage to the environment. Gear these studies to current cropping and management schemes.
4. Continue development of soil and tissue N tests. Integrate these tests with the mass balance, N fertilizer management approach.
5. Evaluate recent developments in equipment design regarding precise timing, distribution, injection, and placement of N fertilizer type, as an aid to minimizing N losses to the environment.
6. Direct nitrification inhibitor and N stabilizer research to provide compounds and formulations geared to specific application and management techniques. Mode of action should be known so that chemical formulation can be made with predictable specificity and efficacy.
7. Tailor N fertilizer management techniques to uniform soil areas rather than whole fields, incorporating appropriate GIS.
8. Continue work to quantify and predict applied and soil N behavior, but center N modeling approaches more on implementation and user application, to ensure proper management decisions.
9. Improve information regarding suitable rotations, cover, and N scavenging crops to alleviate nitrate build-up in soil profiles.
10. Obtain additional information on buffer strip utilization. Current methods use slope as the primary controlling factor, but effects of soil, vegetation, and receiving area require study.
11. Direct more attention to the role of soil fauna (e.g. earthworms arthropods, protozoa) regarding N behavior.
12. Determine both nitrate and bacterial (e.g. coliform) contents in studies involving

pollution of potable waters. This is recommended because infantile methemoglobinemia can be a complication of enteritis (bacterial origin), even when nitrate intake is negligible (Bryson and Boeckman, 1989).

13. Continue efforts to modify plant N metabolic processes through genetic manipulation, to improve N efficiency.
14. Develop furrow irrigation techniques to improve uniformity of water applied and facilitate fertigation management practices.
15. Integrate microbial N cycling, fertilizer management practices, and crop N requirements to improve N use efficiency by synchronizing N availability and crop requirements.
16. Develop dynamic models that will predict nitrate movement as influenced by fertilizer N placement, irrigation application system, and plant root uptake pattern.
17. Obtain additional information on managing nitrate losses associated with tile-drained land.

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## RALEIGH, NC

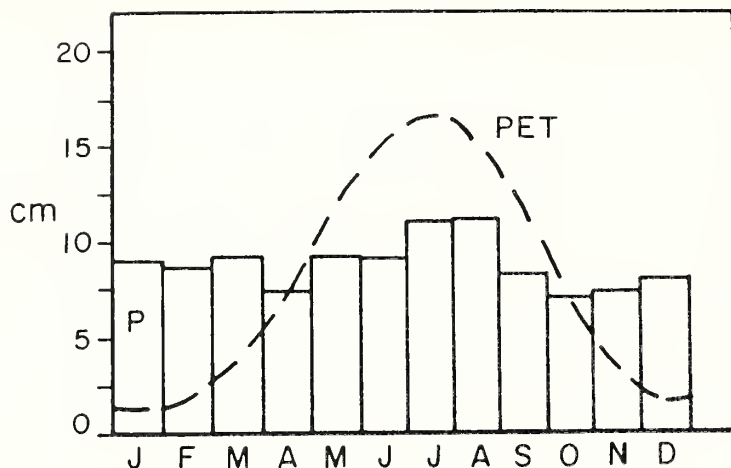


Figure 1. Precipitation (P) and potential evapotranspiration (PET) for Raleigh, NC.

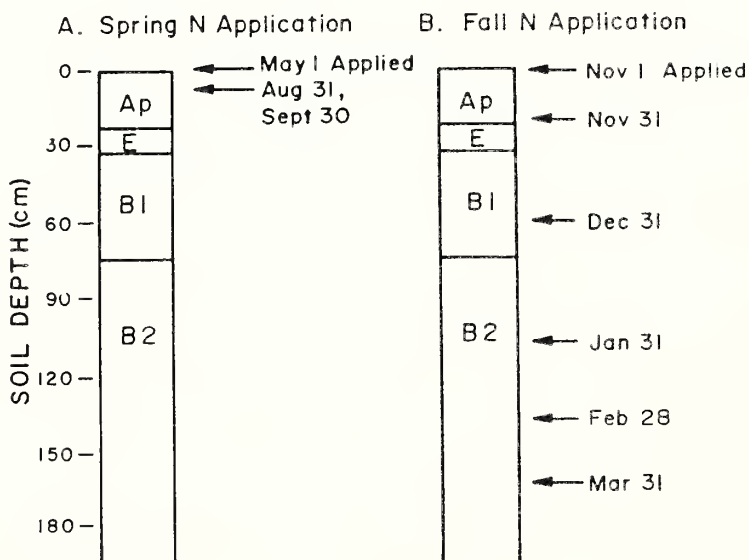


Figure 2. Probable depth of N fertilizer leaching for Norfolk soil at Raleigh, NC.

Table 1. Nitrate contents in soil profiles associated with landuse.<sup>1</sup>

Land Use	No. Soil Profiles	Nitrate-N in .6m soil depth <sup>2</sup>
		(kg ha <sup>-1</sup> )
Alfalfa	13	88
Native grass	17	100
Cultivated drylands	21	290
Irrigated fields <sup>2</sup>	28	562
Feedlots/corrals	47	1594

<sup>1</sup>Study conducted in Colorado (Stewart et al., 1968; Viets, 1975).

<sup>2</sup>Does not include irrigated fields in alfalfa.

<sup>3</sup>Values represent averages for the soil cores.

## ADVANCES IN N USE EFFICIENCY

A. D. Halvorson, B. R. Bock, and J. J. Meisinger<sup>1</sup>

### ABSTRACT

Factors affecting N use efficiency (NUE) by crops, current technology, and future research needs to improve NUE are discussed herein. Long-term trends in corn and wheat yields for the U.S. indicate that yields and rate of N application have increased with time. For wheat, genetic advancements have been credited with 43 to 74% and N fertilization with about 22% of this total increase in yield. Nitrogen use efficiency (g N applied/kg grain) has changed very little since the early 1970's. Thus one may ask the question if any advancement in NUE has really occurred over the last 15 years.

Changes in energy costs and availability, increasing N prices, changes in tillage practices, and environmental concerns have caused a change in attitude of researchers and educators, but apparently not farmers toward NUE in the last 10 years. Urea has become a dominant fertilizer N source in world markets, a phenomenon with implications for NUE. Factors affecting NUE that are currently being emphasized include: N source, timing of N application, placement of N to enhance plant uptake and reduce  $\text{NH}_3$  losses, crop rotation, climatic and soil factors, water supply and crop yield potential, variety/hybrid selection, plant population, disease, weeds, insects, nitrification inhibitors to reduce  $\text{NO}_3\text{-N}$  losses,  $\text{NH}_4$  vs  $\text{NO}_3$  requirements of plants for optimum yield, urease inhibitors

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to reduce  $\text{NH}_3$  losses, irrigation management, and improvements in N recommendations which are based on soil testing for easily mineralizable and residual soil N, credits for legumes, manures, and other wastes applied, and consideration of yield potential based on crop production factors. Factors that affect N uptake by the crop and its yield potential can affect NUE.

Future research needs include development of methods to improve N utilization by crops and reduce N losses to the environment. Development of cropping and crop management systems for efficient water and N use is critical for all crops and climatic conditions. Future research should also address environmental concerns of N in agriculture while maintaining an economically sustainable agriculture in the future. Increasing NUE may have an economic price tag attached for the farmer and society. Our goal should be to integrate all the management, physical, and chemical factors that affect yield into site specific N best management practices that will raise yields with the same amount of N input or maintain current yields with lower N input.

## INTRODUCTION

Most U. S. soils are deficient in N for commercial production of cereals. Nitrogen inputs are therefore needed to sustain commercially viable cereal crop systems. The N inputs for modern crop production can come from manures, fertilizer, or legume residues. Prior to 1950 agriculture relied on manure and legume N inputs from small rotation-livestock systems in order to sustain cereal crop production. Since 1950, use of fertilizer N has increased greatly while wide spread use of manure and legume N has decreased due to the advent of large single species animal enterprises and the costs associated with moving manure. The concentration of animals into large units has increased the manure loading



rates on most livestock farms. Prior to 1975, the primary goal of N management was to improve agronomic effectiveness and economic return. Since 1975, considerable agronomic research has dealt with improving N use efficiency and quantifying N losses from agricultural systems (Hauck, 1984a; Hergert, 1986; The Fertilizer Institute, 1985). Today, improving crop N (manure N, fertilizer N, legume N) utilization in modern agricultural systems is still a major concern (Keeney, 1982, 1985, 1986; Nelson, 1985). Optimizing economic returns and making efficient use of N requires that crop N supplies be adjusted to changes in other crop production factors (Halvorson and Murphy, 1987). Factors that must be considered in optimizing N utilization include: 1) climate and soils; 2) plant-available water and yield potential; 3) tillage method and water conservation practices; 4) timing of N application; 5) N source and placement; 6) soil testing and residual soil NO<sub>3</sub> levels; 7) variety/hybrid selection; 8) plant population and row spacing; 9) seeding date; 10) crop rotation; 11) pests (weeds, insects, disease); 12) efficient irrigation management (minimum NO<sub>3</sub> leaching potential); 13) leaching; and 14) denitrification. Any crop production factor that limits or reduces crop yield with given inputs will generally lower N use efficiency (NUE). Bock (1984) indicated that NUE can be characterized by several relationships: a) the relationship between yield and N rate is yield efficiency (g N applied/kg grain); b) N recovered and N rate is N recovery efficiency (g N uptake/g N applied); and c) N recovered and yield is physiological efficiency (g N uptake/kg grain). Each of these is a distinctly different quantity, so we need to define exactly what is meant by NUE when reporting or utilizing this term. In this paper, we will define NUE as yield efficiency, g N applied/kg grain.

A concern in today's agriculture is the environmental effects of fertilizer

use as well as its impact on food production (Aldrich, 1984). Nitrogen is leaking from agricultural ecosystems into groundwater and surface water (Madison and Brunett, 1985; Chen and Druliner, 1988; Eckhardt and Oaksford, 1988; Keeney, 1985; Keeney, 1986). A balance between food production, profit, and environmental quality must be achieved and maintained by modern agriculture.

An analysis of wheat yield increases relative to advances in technology from 1954 to 1979 shows that wheat yields increased 30 kg/ha per year in the Great Plains states and 43 kg/ha per year in the Cornbelt states (Feyerherm et al., 1988). In the Great

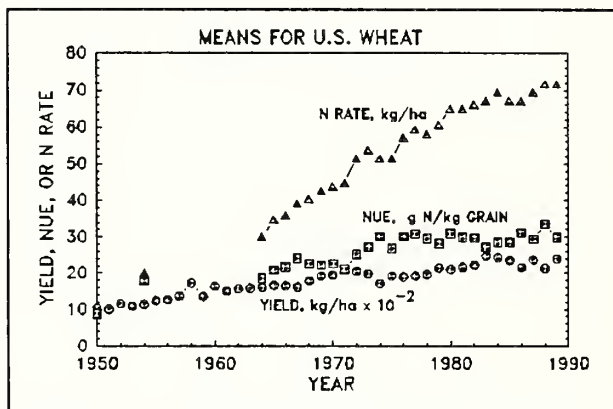


Figure 1. Changes in wheat yields and nitrogen use with time.

increase was attributed to genetic improvement, applied N, and other sources, respectively, compared to 74, 22, and 2% for the Cornbelt. Genetics and applied N accounted for almost all of the yield gain in the Cornbelt, but other factors (irrigation, pesticides, improved tillage, water conservation, etc.)

contributed about one-third of the gain in the Great Plains. Corn and wheat grain yields and N applied to each crop increased from 1954 through 1988 as shown in Figures 1 and 2 (Adams et al., 1958; Hargett et al., 1987; ERS, 1985; NASS 1988). The ratio of fertilizer N applied to grain yield shows that

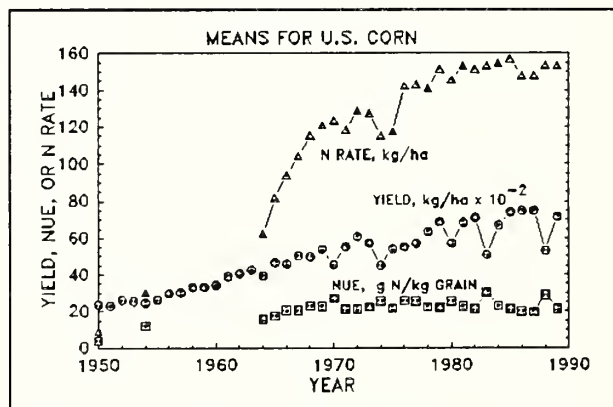


Figure 2. Changes in corn yields and nitrogen use with time.

about 30 g N/kg wheat and about 22 g N/kg corn has been maintained for the past 15 years. This translates into an efficiency of about 55% (kg N in grain/kg N applied) if we assume wheat contains about 2% N, corn about 1.5% N and both grains contain about 15% moisture. This may indicate that on a national basis, little progress has been made in changing NUE by these two crops over the past 15 years. Obviously, NUE can greatly affect N losses from agricultural ecosystems; because N not recovered by the crop potentially may be lost to the environment.

Hauck (1984a) summarized the fate of isotopically labelled N applied to soils. Generally, 50 to 60% of the N applied is taken up by crop plants during the season of application, with a range of 25 to 80%. About 25% of the N applied remains in the soil in inorganic or organic forms after cropping with a range of 15 to 45%. About 25% is lost from the soil-plant system through leaching, denitrification, and/or ammonia volatilization. These numbers represent the partitioning of applied N among plants, soil, waters, and atmosphere during and immediately after the growing season.

Nitrogen application practices, mainly N rate, timing, and placement can have significant effects on NUE. Technological approaches for increasing the efficiency of fertilizer N use by crop plants have included the use of (a) slow-release N fertilizers; (b) chemicals that inhibit biological N transformations in soils; (c) amendments to N fertilizers that alter their physical and/or chemical properties; and (d) improved crop and soil management practices (Hauck, 1984b). These N application practices and technological approaches are directed mainly toward reducing N losses or maintaining an adequate supply of plant-available N in the plant root zone. Major avenues of N loss from the soil-crop systems are leaching, biological denitrification, and ammonia volatilization from

soil.

The topic of N use efficiency is broad and covers many crops and management systems; therefore, our comments will deal mainly with wheat and corn. Recent review articles provide detailed discussions of many of the factors discussed in this paper.

#### FACTORS AFFECTING N USE EFFICIENCY

Nitrogen use efficiency is affected by many factors. Some are not subject to management (weather, soil type, etc.), but many are readily managed (crop variety, N rate, N timing, tillage, irrigation, etc.). Some of the major factors affecting NUE will be discussed.

##### Climate and Soils

Temperature (air and soil), precipitation (amount and distribution), and length of growing season (frost free days) determine crop adaptation and greatly influence yield potential, particularly of dryland crops. Soil physical and chemical characteristics also influence yield potential. Soil texture affects the quantity of soil water that can be held for crop use. Soil compaction can reduce plant growth, root growth and penetration, and water and air movement in soil which can contribute to reduced yields and inefficient use of plant available N. Soil acidity and salinity can decrease crop yield potentials and response to applied N. Soil organic matter content and parent material influence native soil fertility level and plant nutrient supplying power.

##### Yield Potential and Crop N Requirement

The crop N requirement is determined by the type of crop and its yield potential. Predicting yield potential or setting a realistic yield goal is critical for efficient N utilization (Bock and Hergert, 1989). The higher the

yield potential, the greater the total N requirement needed to achieve this yield and maintain crop quality (Black and Bauer, 1986). Wheat generally requires about 40 g N/kg grain to optimize yields (Halvorson et al., 1987), corn about 20-23 g N/kg grain (Fixen, 1985; Meisinger et al., 1985). To optimize profits (least cost per unit of produce), a farmer should neither under nor over estimate a yield goal. Yield limiting factors that can not be controlled by a producer should be considered when establishing yield potentials and determining plant N requirements to insure optimum NUE. For example, yield potential is directly a function of water available for plant growth in semi-arid and arid regions, particularly for dryland crops. In these regions, plant-available water (stored soil water plus growing season precipitation) can be used to predict yield potential (Halvorson and Kresge, 1982; Isfan, 1979).

Nitrogen uptake patterns vary for different crops (Olson and Kurtz, 1982; Bauer et al., 1987). Crops differ in time of year they need N and in total N requirements; however, crops generally need a large amount of N over a relatively short time. Wheat uses about 1.4 kg N/ha (3 lb/a) per day during its "grand (rapid) period of growth" and the corresponding value for corn is about 1.1 kg N/ha (2.5 lb/a) per day. Therefore, in order to insure an adequate N supply in N deficient soils, a rapid N release source or a large pool of  $\text{NO}_3$  from mineralization is needed just before this "grand period of growth".

Much progress has been made in the area of establishing realistic yield goals in the last 10 years (Bock and Hergert, 1989); however, more correlation work is needed in the future to establish yield-N need relationships for crops other than wheat and corn.

#### Soil Testing and Predicting Plant N Requirements

Twenty years ago, soil testing for  $\text{NO}_3$ -N was considered of limited value

in crop production. However, research has shown that  $\text{NO}_3\text{-N}$  tests can be used very effectively for improving N fertilizer recommendations. Crop N requirements have been more clearly defined for many crops as agricultural research has studied crop N uptake associated with various yield levels. Soil  $\text{NO}_3\text{-N}$  tests can increase fertilizer NUE by enabling adjustment of fertilizer N rates to reflect soil  $\text{NO}_3\text{-N}$  content and predicted crop needs. Soil N tests estimating residual  $\text{NO}_3\text{-N}$  within the root-zone (0.6-1.8 m depth) works very well in drier areas of the western U.S. (west of Missouri River). Many western states have active  $\text{NO}_3\text{-N}$  test programs in operation (Hergert, 1987).

Soil  $\text{NO}_3\text{-N}$  testing also is proving to be useful in humid climates, particularly after drought years when residual  $\text{NO}_3\text{-N}$  is likely to be high and after dry winter seasons (Bock and Kelley, 1989; Bundy and Malone, 1988; Mapels et al., 1977). The participants of a soil testing workshop held in February 1989 at TVA emphasized the need for soil and/or plant N tests that can refine N rate recommendations and identify N non-responsive sites as a means of reducing adverse environmental effects of agricultural N and improve economic returns in humid areas of the U.S. (Bock and Kelley, 1989). A pre-sidedress soil  $\text{NO}_3\text{-N}$  test (nitrate measured in the top foot of soil just before sidedressing time) has been highly correlated with corn response to N fertilizer in Vermont (Magdoff et al., 1984), Pennsylvania (Fox and Piekielek, 1984; Fox et al., 1989), and Iowa (Blackmer et al., 1989), areas where soil  $\text{NO}_3\text{-N}$  has been considered impractical in the past. Root-zone sampling for residual  $\text{NO}_3\text{-N}$  provides an excellent opportunity for improving N use efficiency.

Fertilizer recommendations have been adjusted to give credits for other N sources such as manure and legume crops. Determining N rate requirements based on yield potential and N budgets (i.e. - residual soil  $\text{NO}_3$  and  $\text{NH}_4$ ; organic

matter; mineralization capacities, etc.) can greatly enhance NUE. A major goal for the next 10 years is to convince all commercial concerns to adopt soil NO<sub>3</sub>-N testing as a standard part of making N rate recommendations. Obtaining near maximum yields, maximum N use efficiency, and minimal residual soil NO<sub>3</sub>-N carryover at the end of the growing season should be the goal of a proper N fertilization program.

#### Timing of N Application

Timing of N application has received much attention, especially with regard to preventing or reducing the leaching potential of NO<sub>3</sub>-N in agricultural systems (Follett et al., 1989). Application of N just before the time of most rapid N uptake generally assures the best crop use of N under irrigation and in humid areas (Welch, 1971; Meisinger, 1984). In semi-arid areas, lack of rainfall can result in N that is positionally unavailable (in dry surface layers) to the crop at critical growth stages, if the N is not applied early enough. Much of the leaching potential exists between cropping seasons or before crops start growing rapidly. In these situations, post-plant application of N is effective because N is supplied in close proximity with crop N uptake. Data from Malzer and Graff (1984, 1985) shows that less N was required to optimize corn grain yields when sidedressed than when N was applied preplant. In irrigated areas, applying N with irrigation water just preceding maximum N uptake periods is another good method for improving N use efficiency. Careful irrigation management must be practiced in order to avoid over-irrigation, resulting in movement of NO<sub>3</sub> out of the root-zone. Another problem is untimely rainfalls which occur just after "fertigation". Fertigation is the application of N via an irrigation system by injecting the N into the water flowing through the system. Irrigation scheduling is being adapted to improve water use efficiency which should improve NUE by

reducing leaching losses. Gardner and Roth (1984) present a thorough discussion of irrigation method as related to N application.

Split N applications for corn/wheat are known to be the most efficient way to supply N to corn even in drier climates, provided the N is placed into the root-zone and that adequate N is supplied in early part of growing season (Olson et al., 1964; Boswell et al., 1985). With no-tillage or conservation tillage, split N applications have been shown to be especially beneficial due to greater denitrification potential in no-till (wetter soil, more organic matter, denser soil), rapid immobilization of surface applied N in previous crop residues, and greater infiltration of water (with consequent, greater potential leaching) in no-till (Fox and Bandel, 1986; Thomas and Frye, 1984; Wells, 1984). Split applications of N are being evaluated and promoted for use on corn, particularly with irrigation. Split applications of N for dryland wheat are being evaluated as a means of increasing wheat production levels and enhancing grain protein (Alley et al., 1988; personal communications with Armand Bauer, USDA-ARS).

#### N Source and Placement

Consumption of solid fertilizer N sources has changed considerably from 1962 until now (Harre and Bridges, 1988). In 1962, ammonium nitrate, ammonium sulfate, and urea occupied 27, 18, and 5% of the world share of N consumption, respectively. In 1986, the world consumption of these respective products were 15, 5, and 37%. Thus urea is now the dominant N source in world markets. Other N sources, primarily anhydrous ammonia and liquid N sources occupied 29% of the world N market in 1986. In the United States, anhydrous ammonia, N solutions, and urea represent about 40, 20, and 15% of the N fertilizer use. Urea use has increased at the expense of ammonium nitrate.



In summarizing N-source comparison research with respect to N use efficiency and yield response, Hargrove (1988) indicated that when N fertilizers are not incorporated, urea is inferior to ammonium salts as an N source on noncalcareous soils, but for calcareous soils urea can be equal to or only slightly inferior to ammonium nitrate and superior to ammonium sulfate. Fertilizer physical form also affects  $\text{NH}_3$  loss. When broadcast, urea-containing N solutions tend to lose more  $\text{NH}_3$  than dry sources. Thus, with increasing use of urea and urea-containing N solutions, agronomic management practices in the future need to be developed and adapted that will insure high levels of NUE from these N sources. Method of application will greatly influence  $\text{NH}_3$  loss, with maximum losses occurring with broadcast surface applications that receive no incorporation.

Fertigation is one way to reduce N volatilization losses (water carries N into soil), however, some loss may still occur. Fertigation has been practiced with sprinkler, furrow, and drip irrigation (Hargrove, 1988; Randall, 1984). Fertigation also allows timing of N application to coincide with plant N demand periods. Banding N sources on the soil surface or below the soil surface will generally reduce  $\text{NH}_3$  losses, but may or may not increase yields compared to surface broadcast applications, depending on the N loss processes at the specific site.

Bock (1987) reviewed the agronomic differences between supplying plants with  $\text{NO}_3$  and  $\text{NH}_4$ . The relative level of each of these N species in soil can affect crop nutrition, N availability to the roots, and N losses from the root zone. He indicated that inconsistent responses to enhanced ammonium nutrition have been reported from field studies. For corn, the trend is for enhanced ammonium nutrition to increase grain yields of the highest yielding hybrids and

within high yield crop production systems. A potential exists for achieving greater yield responses by selecting genotypes specifically for their ability to respond to enhanced ammonium nutrition. The concept of enhanced ammonium nutrition needs considerable development before it is ready for general use by farmers.

The release characteristics of the N source being utilized should also be considered in relation to the N uptake pattern of the crop being grown. Highest efficiency will be realized if the N release occurs just before (about 2 weeks) the rapid N uptake pattern of the crop. Slow-release commercial N fertilizers are of four types: (1) water soluble N sources with coatings; (2) materials of limited water solubility containing plant-available forms of N; (3) materials of limited water solubility, which, during decomposition release plant-available forms of N; and (4) water-soluble materials that gradually release plant-available N (Allen, 1984; Hauck, 1985). These types of fertilizers represent a wide range in patterns of N release. Nitrogen release patterns differ for various manures. For example, poultry manure has very rapid N release and needs to be used almost like fertilizer N (Bitzer and Sims, 1988; Sims, 1986) while cattle manure has a slow to rapid N release pattern depending on type of manure, bedding, storage conditions and application conditions (surface vs. incorporation). One of the best ways to use manure efficiently is to test it for  $\text{NH}_4\text{-N}$  or total N content, then base application rates on actual analysis and application methods (Klausner and Bouldin, 1983; USDA, 1979). Sludges or composted sludges are also good N sources but the N release patterns can be greatly reduced by composting (O'Keefe et al. 1986; Epstein et al. 1978; Sommers and Giordano, 1984). Loading rates and N use efficiency should therefore be adjusted for N release characteristics of the sludge or manure (Sabey et al.

1975; USDA, 1979). Conventional N fertilizers give virtually immediate (within 2 weeks) N availability since most of these materials contain or convert quickly to  $\text{NH}_4$  or  $\text{NO}_3$ .

For high N use efficiency, ideally the N source should be placed where the N release occurs within the active water uptake portion of the crop's root-zone. Nitrate moves to roots by mass flow with water. In sub-humid and arid parts of the country (west of Missouri River), N sources need to be placed in the root-zone to give best N uptake efficiency and avoid having the  $\text{NO}_3$  accumulate on the dry soil surface where water uptake is low.

Placing N below the soil surface will reduce N volatilization losses, reduce the amount of organic matter tie-up of N, and generally make the N source more readily available to the crop. Advances in fertilizer application equipment will enhance NUE (Murphy and Beaton, 1988; Randall et al., 1985). Combining fertilizer application equipment with computer technology will allow commercial application of prescribed amounts of N for each soil type as the applicator proceeds across a field. Fertilization by soil type should reduce N leaching losses, reduce over and under fertilization problems, and result in improved NUE.

N placement for no-tillage (or conservation tillage) is especially important for urea containing fertilizers. (Fox and Bandel, 1986; Thomas and Frye, 1984; Murphy and Beaton, 1988). Urea fertilizers quickly hydrolyze on the urease rich surface residues of no-till and can lose significant amounts of N through ammonia volatilization (often 5-25% of the urea N). To control these losses, an effort needs to be made to incorporate urea sources by injection behind a coulter, knifed in with common anhydrous equipment or to use N sources not containing urea (Fox and Bandel 1986; Thomas and Frye 1984; Wells, 1984). These same problems also apply to manure N (which contains urea) in no-till

systems. Non-urea N sources generally can be spread on the soil surface with good results. An exception is ammonium sulfate which has a relatively high potential for ammonia volatilization from calcareous soils.

#### N Fertilizer Amendments

Nitrogen use efficiency may also be increased by the use of additives or amendments to N sources. Seven chemicals are produced commercially worldwide for use as nitrification inhibitors with N fertilizers. In 1984, only two chemicals, nitrapyrin and Terrazol, were licensed for use as nitrification inhibitors in the U.S.. Dicyandiamide (DCD) was being test marketed (Hauck, 1984b). The search for urease inhibitors to control rapid urea hydrolysis and the consequent liberation of ammonia is becoming more important as farmers use increasing amounts of urea and UAN solutions, especially with non-incorporated surface applications (Radel et al., 1988; Voss, 1984).

Nitrification inhibitors should be viewed as a N management tool. The benefit to be derived depends on the soil type, time and rate of N application, and weather conditions between N application and crop uptake. The greatest potential for benefits are with soils that frequently remain saturated with water during the early part of the growing season, primarily the poorly drained soils. Coarse-textured soils are likely to benefit more than the finer-textured soils (except those that are poorly drained), since the use of nitrification inhibitors will reduce the high potential for leaching that exists with such soils (Hoeft, 1984).

#### Cropping Systems

Rotation of crops within a cropping system often shows beneficial effects on crop yield (Pierce and Rice, 1988; Power and Doran, 1988). Higher yields resulting from rotating crops at the same N level will improve NUE. Hook and

Guscho (1988) point out that with multiple cropping systems, N that is not utilized by the first crop can often be utilized by the second crop (providing allowances are made for residual soil N), thus giving more efficient use of N. Jones et al. (1980) found that NUE was higher for annual cropping systems than with a wheat-fallow rotation. Utilization of flexible, more intensive cropping systems to make more efficient use of water will also make more efficient use of N inputs. Cropping systems designed to utilize water efficiently can reduce the loss of N through leaching (Halvorson and Kresge, 1982; Halvorson, 1988). Using cropping systems with legumes (soybeans, forages, etc.) is also an excellent way to improve recovery of residual N since legumes generally utilize available soil N before they begin to fix N (Meisinger and Randall, 1989). Cropping system design needs to consider crop water use patterns, rooting depths, and N needs.

#### Variety/Hybrid Selection

Variety or hybrid selection for specific site conditions can greatly influence yield potential, response to N, and profitability. A variety/ hybrid's response to the environment affects both yield and crop quality. Selecting the best adapted varieties or hybrids with the best yield potential for a given area will enhance NUE efficiency for a given N rate. When N rate is adjusted in accordance with yield potential, varieties/hybrids with the highest yield potential may or may not give the highest NUE. Varieties should be chosen for resistance to lodging, diseases, and insects under high N fertility, high yield environments and/or sensitivity to low soil pH conditions. Timian and McMullen (1986) found the spring wheat variety "Olso" had a 33% reduction in grain yield when infected with the wheat streak mosaic virus versus a 98% reduction for "Olaf". Unruh and Whitney (1986) reported that the winter wheat varieties

"Newton" and "Tam 105" were extremely sensitive to low soil pH, high Al concentrations, whereas "Hawk" and "Bounty 203" hybrid showed more tolerance, with yields 3 to 4 times greater. Grain yields are also influenced by plant population and row spacings (Alley et al., 1988).

### Economics

From an individual field cost-price economic stand point, generally it is better to error on the high side than on the low side of N recommendations with current N:crop price ratios (Bock and Hergert, 1989) because under dryland, N not taken up or lost in dry years tend to remain as residual N for the next crop, depending on winter leaching. This is particularly true if N limits yields in wet years; therefore, to error on high side of N recommendations is more economical, but not as environmentally sound. Thus, the immediate short-term economics of the farmer are potentially out of harmony with the long-term environmental goal of minimizing NO<sub>3</sub> losses.

In recent years, there has been considerable interest in the concept of Maximum Economic Yield (MEY) (Wagner, 1988). The MEY concept can enhance or improve NUE by encouraging farmers to use best management practices (BMP) to achieve optimum yields with the most profit. The objective of MEY is to obtain the most economical yield return (not maximum yield); therefore, application of excessive N from an economical perspective is not encouraged. Soil testing, planting date, plant population, herbicide, fungicides, harvest, varieties, etc, are all factors that will improve yield and profit potential (Alley et al., 1988). However, economic and environmental goals relating to NUE may not always be totally compatible (Bock, 1984). The resolution of this situation will likely involve the farmer improving NUE by soil testing, realistic yield goals, etc. and society realizing that it must accept some NO<sub>3</sub> loss to the environment in

deeply rooted crops to appropriate soils.

Although genetic variability in several root characteristics has been demonstrated (O'Toole and Bland, 1987), apparently little effort has been expended in using this information to predict genotypic effects on the fate of  $\text{NO}_3^-$ -N under different conditions with deeply rooted species. Recent ARS work at Blackland and St. Paul are exceptions to this. Some knowledge of root distribution among cultivars of less deeply rooted species like wheat has been gained (Comfort et al., 1988). Development of minirhizotron technology (e.g., Upchurch and Ritchie, 1983; Pettygrove et al., 1988; Box et al., 1989) may greatly expand our knowledge of root extension, distribution, and senescence. If results with winter wheat (Weir and Barraclough, 1986) hold with other species, then water inflow (and presumably  $\text{NO}_3^-$ -N uptake) is maximized in only one depth increment at a time and depends strongly on soil water availability. Therefore, descriptions of root length density and depth distribution alone will not suffice to predict  $\text{NO}_3^-$ -N uptake patterns.

Potential disadvantages of taprooted,  $\text{N}_2$ -fixing species include:

1) the presence of abundant effective nodules on roots at depths sometimes exceeding 4 m (Fox and Lipps, 1955; Virginia et al., 1986), which implies that N deposition can occur far below typical rooting depths of many species, if N loss from nodules (Ta et al., 1986) or frequent nodule senescence occurs; and, 2) the formation of deep, continuous macropores, which can have a disproportionate influence on movement of water through the soil because of by-pass flow (Beven and Germann, 1982). The first process may be important only in specific situations (e.g., presence of a water table at the base of the root zone, as in Fox and Lipps, 1955). Presence of deep, continuous macropores may be beneficial to ground water quality by promoting flow of uncontaminated rain or irrigation water, but

can adversely affect quality when recently added contaminants (manure, residues, pesticides) are moved by preferential water flow. The extent of interaction between water in macropores and the soil matrix depends to a large degree on antecedent soil moisture (Scotter and Kanchanasut, 1981).

Methods to recover residual soil  $\text{NO}_3^-$ -N are needed, both as standard practices in conditions highly subject to leaching (e.g., irrigated sands) and as emergency procedures for use when excessive amounts of  $\text{NO}_3^-$ -N have moved below the root zone of typical crops. Selection and improvement of grass and legume species is required for their use as cover crops, intercrops, and catch crops. The diversity of planned uses requires that selection be made for different characteristics, such as symbiotic  $\text{N}_2$ -fixation capacity, shade tolerance, and rapidity of root extension and shoot growth at low temperatures, respectively.

#### Crop Cultivars

Crop cultivars can vary significantly in N accumulation patterns (Johnson et al., 1967; Rhoades and Stanley, 1981) and may therefore be selected for specific crop production systems (De and Singh, 1981). However, there is continuing controversy about the presence and importance of different N accumulation patterns among crop cultivars. Much of the disparity in results can be attributed to varying environmental conditions (Beauchamp et al., 1976) or sources of N supply (Randall et al., 1987). When soil water and/or N supplies are limited, corn hybrids tend to behave similarly (Randall et al., 1987). Several of the presentations in the recent symposium "Future Developments in Soil Science Research" stressed the increasing role crop genotypes differing in water and N uptake patterns will have in future cropping systems (Boersma, 1987).



Genotypic differences in N uptake patterns may be related to the ability to rapidly accumulate N when it is available in excess of crop requirements (Millard, 1988), to store N in reusable forms even when N supplies limit growth (Millard, 1988), or simply to the continued ability to absorb N as it becomes available (Russelle et al., 1983). Because selection for N uptake efficiency under high N supply may not necessarily result in improved physiological NUE (Moll et al., 1982), simultaneous improvement in both N uptake and N utilization will require development of effective selection procedures under one or more fertility environments.

Perhaps the concept of "N conserving" cereal genotypes will become more relevant in the future. These would be genotypes which are efficient in absorbing N and in producing grain yield, but which retain large proportions of absorbed N in non-harvested vegetative parts and roots to be recycled in the field (Moll et al., 1982). Ideally, genotypes could be selected for their ability to absorb N rapidly whenever it is present during the growing season, perhaps with improved ability to absorb ammonium-N as it becomes available (Bloom and Smart, 1987), but which do not experience significant yield losses when N supply is temporarily limited. Relative N uptake efficiency for a wide range of cultivars and crops is not known.

### Modelling

Significant advances have been made in the development of simulation models to predict the dynamics of N transformation, plant uptake, and losses in natural ecosystems (Reuss and Innis, 1977; Parton et al., 1988), pastures (Kirchmann et al., 1988), crop rotations (e.g., Gustafson, 1988), and monocrops subjected to differing tillage systems

(e.g., Shaffer and Larson, 1982) or irrigation management (e.g., Watts and Martin, 1981). Improvement, integration, and validation of these models for important agricultural ecosystems will highlight areas of knowledge deficit and will be useful in predicting appropriate cropping systems for particular conditions. Extending the use of these models to groups like the SCS, as is being done with EPIC (Putman et al., 1989), will provide a good opportunity for model verification. Watershed-scale models, such as AGNPS (Young et al., 1989), are being developed or improved at at least six ARS locations.

It is critical that the process-based models be formulated with accurate data and biologically-relevant relationships (Van Veen et al., 1981). More detailed information is needed on N mineralization, the influence of roots on water and solute flow, and water interception and redirection by crop canopies. It is surprising that the importance of the last topic has been appreciated and widely studied in forestry, recognized in corn and soybeans for a century (Wollny, 1890), more recently highlighted in potatoes (Saffinga et al., 1976; Jefferies and MacKerron, 1985) and pastures (Kanchanasut and Scotter, 1982), but not intensively studied in most agricultural systems (Clothier, 1988). The importance of stemflow in closely-spaced monocots probably varies with canopy architecture (compare Kanchanasut and Scotter, 1982, with Butler and Huband, 1985). The probable impact of nonhomogeneous water input on subsequent processes in soil (solute flow, N mineralization, denitrification, etc.) seems obvious and requires our serious attention.

A recent conceptual model attributes many of the effects of various cropping systems on soil structure to the creation and destruction of soil macropores (Gibbs and Reid, 1988). The applicability of this model to various climatic and edaphic regions should be tested; fundamental

data are needed which describe effects of root longevity of different species, earthworm burrowing, and soil shrinkage on macropore number, size, and stability.

The rate and regulation of N mineralization during and after crop growth must be better understood to promote improved organic-N source management (Doran et al., 1987). There is disagreement about the characteristics which regulate plant litter decomposition (compare Müller et al., 1988, with Frankenberger and Abdelmagid, 1985). There is also considerable controversy about the methods used to determine potentially mineralizable N (e.g., El-Haris et al., 1983; Beauchamp et al., 1986; Cabrera and Kissel, 1988a and 1988b). In most of our efforts to measure N mineralization processes, we have mistakenly concluded that the only important pathway of N mineralization is through bacteria, and have developed aerobic incubation techniques optimized for bacterial growth. Clarholm (1985) concluded that bacterial mineralization of N has been underestimated, because these methods have routinely ignored the need for suitable C substrates to stimulate microbial growth (Lynch, 1976). Assumptions used in modeling N transformations influence predicted results (Hadas et al., 1987). In addition, net release of N from bacteria requires an alteration in environment or the presence of higher members of the food chain, such as protozoa, fungi, and nematodes (Elliott et al., 1979; Clarholm, 1985; Ingham et al., 1985). Nitrogen accumulated in the rhizosphere of alfalfa (Lory et al., 1988; Russelle et al., 1989) or grasses (Biondini et al., 1988) may represent a readily mineralizable fraction that is also strategically located for absorption by roots of the subsequent crop. These results may help explain why relative differences in N mineralization potential among cropping systems do not always account for measured N availability (e.g., El-Haris et al., 1983; Thicke

and Russelle, 1984). The impact of earthworm activity on nutrient fluxes is not understood and is spatially and temporally complex (Lavelle, 1988). These results and the failure of specific biocide treatments to reduce nutrient availability (Wright and Coleman, 1988) emphasize the need for further research on the role soil invertebrates play in mineralization processes. Work by Woods et al. (1987) is one of the first to incorporate these concepts.

Recent ARS research has demonstrated the potential of heat units (growing degree days) for predicting C and N mineralization from organic materials (Honeycutt et al., 1988). This approach may provide consistency between laboratory measurements and field results, and furthermore could be used directly by farmers.

We also have an incomplete understanding of the influence of living and decaying plant roots on denitrification (Smith and Tiedje, 1979; Haider et al., 1985), and recent work by Parkin (1987) and Klemedtsson et al. (1987) emphasizes the need to learn more about these processes. Information is needed on the influence of root exudates, root debris, and aboveground litter during crop growth and degradation rates of tissues and indirect effects (such as soil porosity) after crop growth. All of these factors are influenced by cropping system. Finally, there appears to be substantial transformation of N in cold soils (Heaney and Nyborg, 1988). Further investigation will clarify the importance and potential for management of these overwinter processes.

### Conclusions and General Recommendations

Optimum economic fertilizer-N rates are usually below those required for maximum yield (Blackmer and Meisinger, 1989), depending on the relationship between N cost and crop value, except in those cases where crop

quality is best at higher N rates (Parr, 1973). Nitrogen use efficiency is usually inversely related to N application rate (Russelle et al., 1981), at least below maximum yield (Fried, 1978). Therefore, losses of  $\text{NO}_3^-$ -N should be reduced by limiting fertilizer-N applications to economically optimum levels (Bock, 1982). This same concept applies to other sources of N, such as legume residues (Hesterman et al., 1987) and manures. Selection of legume genotypes that deposit large amounts of N in the soil may be advisable for intercropping or mixed cropping systems, but inadvisable for rotations of particular monocrops or in warm, humid climates. Incorporation of large quantities of low C:N crop residue may be inherently inefficient because N mineralization rates can exceed crop N uptake (Smith et al., 1987), whereas similar residues may be mineralized too slowly when left on the soil surface (Huntington et al., 1985). Synchrony of cereal crop N requirements with N mineralization from high C:N residues is achieved under some conditions (Waggar et al., 1985), but not others (Power et al., 1986). There is a pressing need to understand the process of N mineralization from soil organic matter and plant residues. Current efforts through the WEP project to predict residence time of crop residues on the soil surface require an understanding of residue decomposition. If these efforts were coordinated with those relating to N release from residues, more rapid progress could be expected.

A strategy for reducing the potential for  $\text{NO}_3^-$ -N leaching into ground water was outlined by Papendick et al. (1987). The proposed strategy includes: 1) improved fertilizer-N management to more closely match inorganic N supply with crop needs and to avoid over-fertilization; 2) reduced fertilizer-N additions by alternating crops with low and high soil-N requirements and using legumes, animal manures, crop residues, and other organic wastes; and, 3) reduced residual soil  $\text{NO}_3^-$ -N by using

cover crops and N-scavenging crops in rotation, and alternating shallowly and deeply rooted crops. Earlier, Olsen et al. (1970) had concluded that  $\text{NO}_3^-$ -N losses could be minimized by reducing the frequency and area of N-fertilized crops grown in rotation (e.g., by increasing the proportion of legumes in the cropping system), limiting fertilizer-N rates, avoiding excessive irrigation, and maximizing the amount of time crops are actively growing. Clearly, use of residual soil  $\text{NO}_3^-$ -N information gathered from soil testing and choice of appropriate yield goals can influence fertilizer-N recommendations (Schepers et al., 1986). Our analysis of the literature generally supports these recommendations, with the addition that multiple cropping and intercropping should be considered where feasible, and with reservations concerning the claim that including more animal manures or  $\text{N}_2$ -fixing crops will necessarily reduce  $\text{NO}_3^-$ -N losses.

Addiscott (1988) reviewed long-term drainage water data from Rothamsted plots and fit  $\text{NO}_3^-$ -N loss data to an exponential decay curve, adjusted for precipitation. He concluded that research should attempt to control N losses from organic matter decomposition during fall, rather than emphasizing only fertilizer-N management. He suggested that  $\text{NO}_3^-$ -N losses might be achieved through use of catch crops or incorporation of high C:N residues in fall. Contrary to most other recommendations, he stressed that accumulation of readily decomposable organic matter should be avoided to limit production of  $\text{NO}_3^-$ -N during periods of little crop growth. This thesis needs to be evaluated, but may only be applicable to highly controlled systems in which small, infrequent doses of fertilizer-N can be applied to match crop needs.

There is a critical need for comprehensive experiments to test these strategies because each management option has the potential for both

positive and negative impact. For example, decreasing surface runoff by conservation tillage systems generally causes increasing potential for leaching (Sharpley et al., 1987). The ultimate effects of conservation tillage systems and riparian areas on the fate of water and N in various edaphic and climatic combinations must be known to help prevent unexpected and undesirable N losses by particularly sensitive pathways (Follett et al., 1987). Several ARS groups have been and continue working on conservation tillage systems and two groups are emphasizing the impact of riparian areas.

Emphasis must be placed on the space- and time-dependence of leaching events, that is, on position in the landscape and transient water flow in structured soils. Recent work by Kung (1988) is an excellent example of how important nonuniform flow in soils can be. The funnelling he observed in stratified sand could be expected in finer-textured soils with impermeable inclusions, such as textural discontinuities, as well as in the more generally recognized conditions of structured or fractured soils or when macropores are present. Kung's results make clear why past and present sampling practices of soil cores and soil solution suction samples are probably inadequate and, worse, inaccurate for estimating leaching of solutes, and therefore why models based on these data may be misleading. Methods for use of ground-penetrating radar on a field scale and computer-aided tomology (CAT) scanning technology on a "pedon" scale are being developed to visualize soil structure and macropore morphology in situ. After nonuniform water flow in soils is better understood, the influence of different cropping patterns, species, and systems must be investigated.

Increased productivity results in decreased NUE (Floate, 1987). We have the capability to alter the relative importance of different N loss

pathways; indeed, the need for our present concern about ground and surface water quality demonstrates this capability. Intensive agriculture may not be compatible with protection of ground water in some areas (Simon et al, 1988). Improved knowledge of the pathways in the N cycle will strengthen our ability to balance and manipulate N inputs and outflows. An ideal solution might be to promote complete denitrification (to  $N_2$ ) of all  $NO_3^-$ -N that has moved or is likely to move below the rooting zone of desired crops. However, predictable fine-tuning of N management may be impossible because of its sensitivity to weather. Granatstein et al. (1987) proposed that maximum nutrient use efficiency would be achieved by growth of a large microbial biomass when crops are not growing, followed by declining biomass with consequent nutrient release during crop growth. This approach requires that N cycling in the microfaunal population is very efficient when crops are not growing, but less efficient when the plants require N. No-tillage management of soils in the Palouse region appeared to support this pattern of microbial growth (Granatstein et al., 1987).

We suggest that the role played by ARS scientists in improving N efficiencies in cropping systems has been appropriate. Adaptation of cropping practices to local conditions ("mesoscale" research) is usually accomplished by AES personnel, who have major responsibilities to solve current problems within their states. ARS scientists should concentrate on the extremes of the research continuum: the microscale, e.g., understanding fundamental aspects of the systems (water flux, regulation of N cycling, germplasm assessment, etc.); and the macroscale, e.g., developing and evaluating innovative cropping systems for broad regions and synthesizing knowledge into stochastic or deterministic models for researchers and for other end-users. Cooperation in research with col-



leagues from other organizations both within and across state boundaries has been frequent and should continue, because of the advantage gained in access to needed expertise and equipment and the resulting improved productivity per unit resource invested. Work is proceeding in other countries on many of the basic problems discussed here; it behooves us to be aware of their current train of scientific thought and to actively cooperate in solving problems of mutual interest.

Economically viable and environmentally sound cropping systems are urgently needed to address public concerns about ground and surface water contamination and to improve the long-term economic stability of our production systems. In all of our attempts, it is crucial that we build on past achievement, that we neither blindly accept dogma nor extrapolate unreasonably, that we develop and use methodologies appropriate in scale, assumptions, and limitations, and that we constructively critique ourselves and each other to keep our science alert.

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## CONTRIBUTIONS FROM NITROGEN RESEARCH

ARS scientists work individually, in teams, and in collaboration with faculty and scientists in land-grant and private universities, other public agencies, and private institutions. Ideas flow freely, and advances in technology and in science owe much to these fruitful associations. What follows is a representative listing of technological and scientific contributions from nitrogen research over the past 12 years in which ARS scientists played a major role. A complete summary is beyond the scope of this report.

A technological contribution is science put to practice solving a problem -- for a producer, for an action agency, for a household consumer, for industry. A scientific contribution is an advance in pushing ahead the frontiers of science; these matter mostly to scientists, but they are the basis of tomorrow's technologies.

For ease of presentation, the contributions are classified for their relevance to major contemporary social issues -- sustainability of agriculture, water quality, and global climate change. Many contributions address more than one issue -- exemplary of the "spillover" of research on one problem to solving another.

### SUSTAINABILITY OF AGRICULTURE

#### Technological Contributions:

- \* Released alfalfa germplasm with increased nitrogen fixation and nitrogen storage for use in crop rotations.
- \* Developed methods to credit legumes with nitrogen provided to a following crop and spare the usage of manufactured fertilizer.
- \* Released non-nitrogen fixing alfalfa germplasm for use in measuring nitrogen fixation of new varieties in commercial breeding programs.
- \* Developed strategies to credit fertilizer recommendations for nitrogen applied in irrigation water.
- \* Patented strains of soybean root nodule bacteria eliciting increased nodulation and nitrogen fixation.
- \* Improved the timing of fertilizer application by adding nitrogen to irrigation water.
- \* Patented a denitrification inhibitor to reduce gaseous losses of nitrogen from fertilizer.
- \* Acquired and preserved an extensive collection of root nodule bacteria that enable legume crops to be self sufficient for nitrogen, and provided these bacteria to the world's inoculum industry.
- \* Devised a multistep selection procedure to enhance the nitrogen fixation of alfalfa in breeding and germplasm enhancement programs.

- \* Patented a method to match superior nitrogen fixing bacteria and highly productive plants.
- \* Fostered the adoption of realistic yield goals as a basis for nitrogen fertilizer recommendations.
- \* Developed equipment for managing crop residues to improve recovery of nitrogen in them.
- \* Reclaimed mine spoils and increased the productivity of eroded lands with legumes and other organic nitrogen sources.

#### Scientific Contributions:

- \* Discovered an alternate nitrogen fixing enzyme system, and developed structurally altered forms of the enzyme.
- \* Developed stable isotope methods to permit first field scale measurements of nitrogen fixation by hay and pasture legumes.
- \* Developed methods to measure plant available nitrogen in soils and the potential nitrogen supply during the growing season.
- \* Determined how nitrogen accumulation of crops varies with stage of development.
- \* Determined how cropping system, crop management, residue management, and tillage method regulate quantity, availability, and use efficiency of nitrogen in the soil.
- \* Quantified the net nitrogen return to soils of legumes growing in rotation with other crops.
- \* Identified the physical, chemical, and biological benefits of organic nitrogen sources to soils.
- \* Discovered nitrogen fertilizer forms and methods of application to reduce volatilization of ammonia.
- \* Developed micrometeorological methods to measure changes in ammonia concentration in the field.
- \* Prepared antibodies, that will be useful for gene isolation, to five plant enzymes essential for nitrogen assimilation.
- \* Measured transfer of nitrogen from legume to grass, and determined its importance to nutrition of grass.
- \* Demonstrated the pathway of assimilation of atmospheric nitrogen in major legumes that metabolize amides.
- \* Generated and characterized soybean germplasm with "supernodulation" traits that is able to nodulate under high nitrate concentrations.

- \* Developed plant-rhizobia-mycorrhizae-rhizobacteria inoculum procedures to enhance plant productivity.
- \* Generated soybean germplasm having mutations for the key nitrate-assimilating enzyme.
- \* Characterized the genetic, biochemical, and molecular control of plant controlled mutants for nodulation and nitrogen fixation.
- \* Mapped the relationships between genes for nodulation and for nitrogen fixation in soybean root nodule bacteria.
- \* Identified the major transport form of nitrogen in beans and cowpeas, and devised a method of testing this compound in xylem sap to measure nitrogen fixation.
- \* Demonstrated the importance of carbon fixation and conservation in the legume root nodule to nitrogen fixation and transport.

## WATER QUALITY

### Technological Contributions:

- \* Developed prediction and loading models to determine losses of soluble and sediment-borne nitrogen under various cropping practices.
- \* Devised, evaluated, and implemented nitrate tissue tests for sugar beets and potatoes, and implemented a "June" soil nitrogen test for assessment of supplemental fertilizer needs for rest of season.
- \* Developed, evaluated, and implemented use of buffer strips to reduce soluble and sediment-borne nitrogen additions to water.
- \* Developed irrigation scheduling procedures that minimize runoff and leaching of applied water and soluble nitrogen.
- \* Utilized "spoke" injection equipment and point injection techniques to apply fertilizer nitrogen at critical crop growth stages, to increase crop yield from fertilizer applied, and to reduce leaching losses.
- \* Developed and patented a slow-release nitrification inhibitor.
- \* Develop strategies to increase nitrogen use efficiency from animal manures.
- \* Developed strategies to safely apply sewage sludge to land, which became the basis of federal regulations.
- \* Developed systems of managing nitrogen rich crop residues including cover crops and green manures to minimize off-season leaching of nitrogen.
- \* Conceived and developed models to identify sites vulnerable to ground water quality problems and identify nitrogen management strategies to avert risk.



- \* Participated in development and verification of soil tests for nitrogen use on major crops, and of techniques for efficient timing and placement of nitrogen fertilizers.
- \* Determined how tillage practices modify soil environment and nitrification.

#### Scientific Contributions:

- \* Characterized and quantified nitrogen and sediment loads from agricultural watersheds by model development and validation.
- \* Defined principles and characterized concepts of nitrogen transformations used to develop models and expert systems.
- \* Characterized transformations of nitrogen in riparian zones.
- \* Conceptualized the processes involved in preferential flow of water through soils.
- \* Discovered how soil tillage and crop residue management practices mediate soil microbial activity to control nitrogen transformations.
- \* Determined how nitrogen management strategies affect nitrogen retention and losses in watersheds.
- \* Developed methods to measure denitrification in the laboratory and field.
- \* Determined how soil type affects the potential for nitrogen mineralization.
- \* Conceived and developed models to predict nitrification, mineralization, and immobilization of nitrogen in soils.
- \* Determined how soil temperature and water availability affect nitrification.
- \* Identified the primary rate limiting processes in nitrate uptake from soils by crops, and in crop nitrate assimilation.

### **GLOBAL CLIMATE CHANGE**

#### Technological Contribution:

- \* Developed models of crop growth to predict how crop yields and quality respond to possible environmental stresses and climatic scenarios.

#### Scientific Contributions:

- \* Developed methods to measure concentrations of individual oxides of nitrogen under field conditions.
- \* Devised techniques to quantify denitrification in the field.

- \* Conceived approaches to determine the effects of tropospheric ozone on nitrogen accumulation and assimilate partitioning of crops.
- \* Assessed the significance of production of oxides of nitrogen from ammoniacal fertilizers.
- \* Identified role of time and space variation in soil water and available carbon in nitrogen losses to denitrification.
- \* Identified chemical reactions that regulate emissions of nitrogen oxides from soils.
- \* Simulated, with models, denitrification from various agricultural cropping systems.
- \* Demonstrated that nitrogen fixation is one of the first plant processes impaired by stress of the type likely to be associated with climate change.



## RESEARCH IMPERATIVES AND TASKS

As the first step in identifying research needs, each of the six Subject Matter Working Groups defined a problem statement and specific research objectives associated with the problem. Collectively, the six groups identified 31 research objectives, or tasks, of scientific priority. Next, the 31 research tasks were ranked for relative importance by each of the five Crop Management System Working Groups. Finally, each Crop Management System Group ranked the research needs by importance to several societal issues, e.g., sustainability of agriculture, water quality, and global climate change. The Organizing Committee compiled a matrix from this exercise to identify and summarize major research themes, or imperatives, of scientific urgency that are relevant to the needs of major crop management systems and sensitive to societal issues. The results of this analysis revealed four research imperatives within which 24 important research tasks could be arrayed: (1) Develop and Use Plant Germplasm with Improved Attributes, (2) Increase Understanding of Soil N Transformation Processes, (3) Conceive, Develop, and Facilitate Adoption of Acceptable N Management Systems, and (4) Integrate and Transfer Knowledge by Simulation and Modeling.

Imperative 1. Develop and Use Plant Germplasm with Improved Attributes.

### Research Tasks

- a) Identify and map plant and bacterial genes controlling uptake, assimilation, partitioning, and utilization of N ( $N_2$ ,  $NO_3^-$ ,  $NH_4^+$ ).
- b) Determine the physiological, biochemical, and molecular regulation of N ( $N_2$ ,  $NO_3^-$ ,  $NH_4^+$ ) assimilation and partitioning at the cellular, organismal, and crop community levels.
- c) Improve host compatibility and capability for  $N_2$  fixation of root nodule bacteria.
- d) Identify germplasm with improved capability for  $N_2$  fixation and soil N use efficiency, and develop strategies to use the new germplasm in plant improvement programs.
- e) Understand the interdependence of plants and rhizosphere organisms such as *Pseudomonas* spp. and VA mycorrhizae, and their interaction with root nodule bacteria.
- f) Determine effects of environmental stresses, especially those anticipated from global climate change, on plant N metabolism.
- g) Develop legume cultivars with special attributes, e.g., ability to reseed, winter-hardiness, efficient water use, and efficient N scavenging, for use as cover crops and intercrops in new cropping systems.

Imperative 2. Increase Understanding of Soil N Transformation Processes.

### Research Tasks

- a) Determine factors controlling mineralization-immobilization processes, and conceive suitable analytical techniques to allow development of

management practices to synchronize soil N availability with crop needs.

- b) Devise analytical methods to measure denitrification, ammonia volatilization, and other gaseous N transformations and determine the factors controlling these processes.
- c) Improve analytical methods to measure nitrification, and determine how to control nitrification and non-exchangable N transformations by inhibitors and by other methods.
- d) Determine the factors that control the accumulation and decomposition of soil organic matter, and their effects upon the soil physical, chemical, and biological environment.
- e) Assess the dynamics and relative importance of gaseous N exchange between the biosphere and atmosphere, and its potential cumulative effects on global warming.

### Imperative 3. Conceive, Develop, and Facilitate Adoption of Acceptable N Management Systems.

#### Research Tasks.

- a) Determine effects of tillage and crop residue management practices on soil environment and N availability, and on subsequent N transformations and fate.
- b) Evaluate how cropping systems (including cover crops) cause changes in the soil environment and subsequent transformations and fate of N.
- c) Understand how soil and fertilizer management practices cause changes in the soil environment and subsequent N transformations, crop uptake, and fate of N.
- d) Evaluate effects of irrigation and drainage systems on changes in soil environment and subsequent N transformations and fate of all N sources.
- e) Conceive and develop suitable soil and plant tissue tests to accurately assess N deficiencies and produce fertilizer N requirements for various soils, cropping systems, and climates.
- f) Develop the means to accurately predict mineralization rate of N in residues from crops, animal manures, and other organic sources when used with various soils, tillage practices, and climates.
- g) Develop management practices that synchronize the availability of N from all sources with crop N requirements to minimize N losses to atmosphere and water.

#### Imperative 4. Integrate and Transfer Knowledge by Simulation and Modeling.

##### Research Tasks

- a) Develop accurate, process-based algorithms that quantitatively describe the effects of soil, crop, and climate on various N transformations.
- b) Incorporate algorithms into computer simulation models to accurately describe the effects of changes in soil environment resulting from management decisions on the fate of N from all sources, and on crop N uptake.
- c) Develop user-friendly computer models suitable for routine use by clientele that outline the consequences of alternative soil and crop management decisions in terms of crop production, net income, soil quality changes, and environmental quality.
- d) Develop computer models incorporating the foregoing algorithms and on-board sensors to change field equipment settings for planting and fertilization as the equipment passes from one soil condition to another in the field.
- e) Field verify the foregoing models and establish mechanisms for transferring the technology to producers, agri-chemical dealers, extension and action agencies, consultants, and other clientele.

Working Groups frequently accompanied a task statement with a listing of specific objectives and methods to use. Space precludes reporting these lengthy lists of objectives and protocols, which in the last analysis are open to the creative choice of individual scientists. Examples of specific objectives appropriate to these imperatives can be found in the appendices, in reports of individual scientists.

The process of discussion, debate, and prioritization reduced from 31 to 24 (a 26% reduction) the number of research tasks initially identified. The imperatives and tasks identify urgent problems to solve and important science to do which, in the collective judgement of the participants, are consistent with the needs of society.

**COOPERATING AGENCY CONTRIBUTIONS**





## Activities in the Agricultural Research Department

The Agricultural Research Department (ARD) conducts research addressing primarily fundamental problems that reflect emerging issues and concerns, while continuing to look for innovative ideas on the cutting edge of agricultural advancement.

The Department's mission is to —

- *Provide leadership and information on the agronomic and environmental effects of fertilizers and other plant nutrients under crop management systems in a wide range of soil and climate conditions, and*
- *To provide leadership and information on environmentally acceptable use or treatment of waste products.*

The Department's objectives are to—

- *Assess soil and plant factors associated with efficient use of nutrients under various cropping systems,*
- *Develop practices for more efficient nutrient use in cropping systems,*
- *Provide agronomic and economic information for the development and evaluation of fertilizers and fertilizer additives under various management systems,*
- *Determine environmental impacts of plant nutrients on water quality, and*
- *Determine the fate of plant nutrients and other elements or compounds in agricultural, industrial, and municipal wastes applied to soil.*

The Department conducts research in a variety of ways, including—

- *In-house research at Muscle Shoals, Alabama,*
- *Cooperative research with universities and other government agencies, and*
- *Joint ventures with the private sector.*

In-house work is conducted in a number of areas, such as—

- *Nitrogen,*
- *Phosphorus,*
- *Secondary and micronutrients,*
- *Soil and crop management,*
- *Environmental,*
- *Waste Management,*
- *Aquaculture, and*
- *Information management and retrieval.*

Cooperative research is conducted to—

- *Complement in-house research,*
- *Broaden the scope of our research, and*
- *Promote information transfer.*

Examples of these activities may be found in this document.

***For More Information***

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Nitrogen Research Needs  
Francis Thicke  
Extension Service - USDA

- . Further test and develop the "presidedress" soil N test, or other procedures (e.g., tissue testing) to more closely assess crop N fertilizer needs.
- . Develop more precise N credits for manures and legumes under various climates and management systems.
- . Assess the potential for N to leach from legumes under various cropping conditions, and develop management practices to minimize leaching.
- . Develop cropping systems using rotations and cover crops that promote efficient N use and minimize the potential for N leaching.
- . Assess the potential for "maximum economic yield" to result in nitrate contamination of groundwater under various climates and soils.
- . Develop decision aids that can be used by extension staff to help farmers select and manage profitable and environmentally sound production practices.

Barbara M. Vining  
Soil Conservation Service

SCS-ARS-MOU

1. Each participant here should have a copy of the most recent SCS-ARS-MOU. I can supply this to Gary for the mailout with workgroup results.

TOOLS FOR TECHNOLOGY TRANSFER

2. SCS, as well as CES, can provide the link for technology transfer from ARS to farmers. SCS tailors ARS research into usable tools and products which our conservation planners can use.

CLEAR CUT DEFINITIONS OF TILLAGE SYSTEMS

3. There is a need in the scientific literature to fully define what is meant by no-till, conservation tillage, etc. No-till is not instituted year by year. It is continual over years. Some instances in literature have occurred where the no-till plots have been plowed in the fall and initiated in the Spring. This is not no-till.

SCS-ARS LINK

4. SCS can supply ARS with ideas of local or regionalized research needs, since we work with the farmer.

CONSERVATION PRACTICES

5. ARS could assist SCS in continuing research related to current SCS conservation practices used in conservation planning and also ARS can introduce innovative systems then let SCS know the research results.
6. Many scientific investigations have not been run long enough. The literature is full of one and two year experiments. Research needs to be conducted more long-ranged (5-10 years).

RESOURCE MANAGEMENT SYSTEMS

7. SCS tries to plan conservation planning based on resource management systems. Therefore, our needs from research are focused on the effects of whole systems and not necessarily on components. Interactions can be too complex sometimes to single out individual practice effects.

SCS-ARS UPDATE

8. There is a real need of the SCS technical centers to be current in ARS literature. If a mechanism does not exist to transmit information on a periodic basis, one should be instituted. This is best done on a regional basis.

### DATA DISSEMINATION

9. There is a lot of data residing in filing cabinets of scientists which has not been published, but which has great application to current problems. There is a need to get the data in print, even if only in a nontechnical journal or magazine.

### REGIONAL NEEDS

10. There is a need for more applied research which is related to regional agriculture industry needs. Example: The poultry industry in Missouri produces tremendous amounts of animal wastes with high N and P concentrations. Research in the management of large-scale wastes is needed.

### URBAN PROBLEMS

11. Urban effects of excessive N fertilizing of lawns is sorely absent in the literature. How much N is leached in urban areas?
12. Consider the design of systems which control N form-lagoons, impoundment structures, effects on denitrification, etc.

### LANDSCAPE EFFECTS

13. Topo sequences within a microclimate - are conservation effects the same under different topographical sequences.

I realize that pure research is as necessary as applied. I am slanting my ideas (opinions) towards the applied side since this is my work.

## APPENDICES



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**REPORTS OF INDIVIDUAL SCIENTISTS**



**WORKING GROUP 1**  
**SOIL NITROGEN TRANSFORMATIONS**



## L. F. Elliott: Current research

Soil nutrient retention as affected by residue management, crop rotation, cover crops and the soil microbial biomass. Water management practices that minimize chemical backing. Cropping practices that maximize rhizosphere relationships for optimum plant growth. The effect of cropping practices on the soil borne disease organism (Pythium sp.) responsible for carrot forking and the possibility for biological control (Mike Davis, U. C. Davis, P. I.). Crop residue management for optimum crop growth.

## ARS Research Needs

1. Preventing root zone escape of chemical through the use of crop rotations, tillage and residue management, and water management.
2. Integrating current knowledge into cropping systems that are sustainable, environmentally friendly, economic, and user acceptable.
3. Designing tillage and residue management systems that optimize rhizosphere relationships.
4. Continued efforts on biological control of pests.

## CURRENT RESEARCH ACTIVITIES

Burl Meek  
Soil and Water Management Research Unit  
Kimberly, Idaho

OBJECTIVE: To evaluate the effect of cropping systems, conservation tillage, fertilization, and irrigation on nitrogen transformations and nitrate leaching below the root zone.

### Summer 1989:

Determine water and nitrogen flux below the root zone of a crop of sugar beets where various rates of N and water have been applied. The internal drainage method of measuring water flux will be evaluated. Ceramic soil solution samples will be compared to soil sampling as a method to determine nitrate concentration in the soil solution.

### Planning for 1990:

An experiment is being planned to evaluate the effect of cropping sequence, conservation tillage, fertilization and irrigation on nitrogen transformations and nitrate leaching below the root zone.

Systems will be evaluated in terms of:

- (1) Nitrogen flux below the root zone.
- (2) Water flux below the root zone.
- (3) Nitrogen availability to crops from the soil.
- (4) Nitrogen uptake of crops.

The goal of the research will be to match as close as possible the N uptake of the crops to the mineralization from the soil and reduce nitrogen fertilization when it can be done without reducing yield.



## FUTURE PROBLEMS AND RESEARCH PRIORITIES FOR ARS

- (1) Develop simple analytical procedures and models to predict mineralization of soil nitrogen.
- (2) Develop and test methods to measure the nitrate flux below the root zone.
- (3) Evaluate the effect of reduced or no-till on nitrogen transformations.
- (4) Develop methods to evaluate the contribution of macropores to water flow in soils.

Burl Meek  
Soil and Water Management Research Unit  
Kimberly, Idaho

## CURRENT RESEARCH OBJECTIVES

Michael P. Russelle  
USDFRC, St. Paul, MN

Overall objectives of present research are to quantify the accretion, cycling, and loss of symbiotically-fixed  $N_2$  (SFN) in soil-plant systems and to develop management approaches to minimize losses to ground water.

1. Determine the distribution of SFN during growth of alfalfa in shoots, roots, and soil.
2. Determine amounts of SFN lost by leaching during alfalfa growth under field and greenhouse conditions.
3. Determine spatial and temporal changes in nutrient absorption activity of legume and grass roots.
4. Evaluate the influence of macropores on potential nitrate movement during simulated spring thaw in soil under alfalfa or corn.
5. Assess the fate and actual availability of N from decomposing alfalfa residues to succeeding crops of sorghum sudangrass.
6. Test the hypothesis that recovery of legume residue N can be optimized by combining appropriate corn hybrids, tillage management, and N fertilizer rate.
7. Compare the effectiveness of various deeply-rooted forage species in absorbing accumulations of nitrate in the subsoil.

The following are research objectives which ARS is particularly well suited to achieve in the next 15 years, given sufficient resources:

1. Achieve fundamental understanding of food web relationships in N mineralization and immobilization in soil throughout the year, including development of agronomically relevant N mineralization indexes.
2. Achieve fundamental understanding of the factors regulating N availability from decomposing plant tissues and other organic materials, so that N release can be synchronized with N need of the subsequent crop.
3. Select and assess various legume crops for crop rotations and develop economically viable and environmentally sound rotation management systems.
4. Fully assess the input and fate of symbiotically-fixed N in a variety of agricultural ecosystems.
5. Determine limitations to N use efficiency and measure the extent of N losses in forages managed as pasture or hay/ensilage under irrigated and rainfed conditions, including effects on soil physical properties.
6. Select and assess weed and crop species as cover crops and further develop comprehensive and effective management systems for major agricultural regions.
7. Develop methods to assess and genetically manipulate root morphology, root growth, and patterns of N and water uptake of various species to be used for specific roles in cropping systems, including deeply rooted species to remove  $\text{NO}_3^-$ -N from the subsoil.
8. Improve understanding of the direct and indirect effects of roots, root exudates, herbage decomposition, and other regulating factors on denitrification.
9. Improve understanding of nonhomogeneous water input to and movement through soils on subsequent N cycling processes and on the space- and time-dependence of leaching events, with application to structured soils and various plant populations, including riparian areas.
10. Develop practical methods to finely tune N management in various cropping systems, with emphasis on those systems with greatest potential for negative environmental impact.
11. Improve, integrate, and validate research- and "user"-level simulation models and expert systems for important agricultural ecosystems.

## ARS Nitrogen Working Conference

### Future Problems and Research Priorities for ARS

Jean D. Reeder, Soil Scientist, Fort Collins CO

#### General Goal Statements for Future ARS Research:

- A. In the past, agricultural research has focused on increasing crop production. Research was devoted to developing fertilizer and pest control technologies, breeding higher-yielding crop varieties, and developing cultural practices e.g. irrigation and monoculture management practices to enhance plant growth. Agricultural research has led to the development of intensive, industrialized agriculture which requires high energy and chemical inputs (Odum, 1984).

Given the increasing costs of both energy and pollution, agricultural research in the future should be devoted to optimizing profitability (but not necessarily production) while minimizing the negative environmental effects of agriculture e.g. atmospheric pollution, surface and ground water pollution, and soil degradation.

We need to conduct research to develop management practices that reduce the input and output costs of agriculture. That is, develop management practices that:

1. Decrease soil degradation  
(e.g. limited till and no-till practices that reduce erosion and build, rather than degrade, soil organic matter)
2. Increase nutrient retention and recycling so as to reduce fertilizer inputs and nutrient contamination of surface and ground waters.
3. Increase diversity through multiple and rotational cropping so as to minimize soil degradation and reduce excessive dependence on broad-spectrum pesticides.
4. Reduce wastage of irrigation waters.

- B. There is not enough data that quantify the actual impacts of various agricultural practices on the environment. Nor have we adequately evaluated the economics of the various management practices. We need to conduct research to:

1. Quantify nutrient losses (by wind and water erosion and by leaching) that occur with various management practices.
2. Develop simulation models that adequately describe the soil/plant/water system and can be used to evaluate various management practices in terms of economic returns and environmental impacts. Since wind and water erosion processes are selective in the size of soil particles (aggregates) eroded, such models must adequately describe the association of organic matter and nutrients with the various size particles (aggregates) that make up the soil.

#### References

- Odum, E.P. 1984. Properties of agroecosystems. pp. 5-11. In R. Lowrance et al. (ed.) Agricultural Ecosystems--Unifying Concepts. John Wiley & Sons, N.Y.

## ARS Nitrogen Working Conference

Jean D. Reeder, Soil Scientist, Fort Collins CO

### General Problem Statement for Current Research Activities:

Numerous studies have been conducted to evaluate the effects of cultivation and erosion on soil productivity. However, these studies have generally focused on the properties of whole soils; little is known about the effects of cultivation and erosion on the properties of soil aggregates. We know that soil structure protects soil organic matter (OM) and influences OM turnover and thus soil fertility. However, the physical location of OM in soil (e.g. clay adsorbed or occluded in aggregates) is not well understood. Soil aggregation may be a major factor controlling OM transformations. Thus, a better understanding is needed of the amount and type of OM that is associated with the various size aggregates found in soils. We also need to understand how the association of OM with soil aggregates is affected by soil texture and various agricultural management practices.

### Current Research Activities:

1. Cooperative long term field study with G.E. Schuman (ARS, Cheyenne WY) and R.A. Bowman (ARS, Ft. Collins CO)

#### Objectives:

To evaluate differences in aggregate size distribution and stability, and in the association of organic matter with aggregate size in native rangeland vs. marginal cropland.

To evaluate annual changes in these properties in grasslands reestablished on marginal croplands and in croplands established on newly plowed rangelands.

2. Cooperative field studies with R.A. Bowman (ARS, Ft. Collins CO)

#### Objective:

To evaluate the effects of long- vs. short-term cropping on aggregate size distribution and stability, and on the quantity and quality of organic matter associated with aggregate size fractions.

3. Cooperative studies with D.E. Stott (ARS, West Lafayette IN)

#### Objectives:

To evaluate the effects of various crop rotations and tillage practices on aggregate size distribution and stability, and on the quantity and quality of organic matter associated with aggregate size fractions.

To explore possible relations of above organic matter/aggregate properties with sediment losses during simulated rainfall.

To evaluate the losses of dissolved organic matter and the degree of preferential selection of certain organic matter fractions that are lost during simulated erosive rainfall events on two soils with different textures and orders (Alfisol and Inceptisol).

JEFFREY L. SMITH  
Soil Biochemist  
Land Management and Water  
Conservation Unit  
Pullman, WA 99164-6421

FUTURE PROBLEMS AND RESEARCH PRIORITIES FOR ARS

1. The relationship between soil microbial diversity and the declining productivity of the world's croplands.
2. The role of conservation tillage in groundwater contamination by agricultural chemicals.
3. Agronomic implications of the greenhouse effect on the global nitrogen cycle
4. Biochemical mechanisms of the tie-up and release of nitrogen as related to synchronization of plant uptake and utilization.
5. Develop methods for determining N volatilization losses from crop and soil systems.
6. Develop GIS programs for managing fertilizer N on a landscape basis.

JEFFREY L. SMITH  
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### Current Research Activities

Current nitrogen research in our unit centers upon nitrogen use efficiency. Efficiency studies have been initiated investigating the influence of residue management, crop rotation and one-pass drilling operations on the  $^{15}\text{N}$  uptake and utilization of fertilizer nitrogen. These studies utilize  $^{15}\text{N}$  to trace the fate of nitrogen at fertilizer or tracer levels.

We are currently in the second year of an  $^{15}\text{N}$  efficiency study using hard red spring wheat. This study focuses on fall vs. spring nitrogen fertilization of spring crops. In addition the  $^{15}\text{N}$  experiment is designed to determine the efficiency of foliar applied  $^{15}\text{N}$ .

We have initiated several field studies to assess the potential for groundwater contamination from agricultural chemicals. Using  $^{15}\text{N}$  we are tracing N movement as related to slope steepness. At present we have no information on surface vs. subsurface movement of nutrients in our system. In addition we are assessing long term nitrogen movement by analysis of deep cores taken on grid lines for landscape analysis. This information is being used in a simple evapotranspiration model to estimate the potential for N loss by leaching in soils of the Palouse region.

Other nitrogen related projects include:

- a. Low and high input rotation systems.
- b. Nitrogen contribution from legume rotations.
- c. Nitrogen transformation ( $^{15}\text{N}$ ) in native grass and wheat systems: A comparative analysis.
- d. Nitrogen cycling and mineralization potentials in arid ecosystems.
- e. The effect of elevated  $\text{CO}_2$  levels on nitrogen use efficiency in wheat.

CURRENT NSERL SOIL MICROBIOLOGY/BIOCHEMISTRY RESEARCH PROGRAM

DR. DIANE E. STOTT  
NATIONAL SOIL EROSION RESEARCH LABORATORY  
WEST LAFAYETTE, INDIANA 47906

- A. A major thrust of my group is to examine how various soil organic matter fractions affect soil erodibility and identify classes of compounds that can significantly influence erodibility. Future work will emphasize means of exploiting this information in developing erosion control practices that will protect soil surfaces during critical erosion periods in a cost effective manner.

Research literature shows that near-surface soil shear strength is closely correlated with the amount of soil detached by raindrop impact, and the changes in this relationship between soil types is frequently correlated with organic matter content, suggesting a biological contribution. However, most research on how soil microbes affect soil structure has been done in the context of soil aggregation and aggregating agents. The gap between aggregate formation and how microbes affect soil cohesive properties and soil mechanical strength has never been bridged. Studying a single soil type under several different crop rotations has shown a correlation between polysaccharide content and decreased near-surface soil shear strength. Laboratory work indicates that as an organic compound has an increasing affect on large aggregate formation, the near-surface soil shear strength decreases and soil detachment due to raindrop impact increases, however neither measurement takes infiltration rate changes into account.

While it is known that soil organic matter is lost during an erosion event, little research has been done to determine the type of organic matter lost or if a given fraction is eroded preferentially. In cooperation with Dr. Jean Reeder, we are looking at soil organic matter, carbon, nitrogen and phosphorous losses with eroding sediments. In a companion study, we are looking at the relationships between the bulk soil humus and the associated aggregate fractions and sediment losses during simulated rainfall. The properties emphasized are those that we think may differ in soil cultivated to corn vs. soybean.

- B. The other major research thrust of my group is the decomposition of several major crop residues. We are conducting experiments in order to develop algorithms to predict decomposition of 10 major crop residues given a few climatic and soil factors. A computer decision support system is being developed to aid SCS personnel in predicting how changing climatic and management factors change residue mass and percent surface residue cover. This program will allow the user to do some "what if " projections as an aid to choosing appropriate residue management techniques.



USDA-ARS Working Conference on Nitrogen Research  
Future Direction of Nitrogen Research  
Diane E. Stott, Soil Microbiologist  
National Soil Erosion Research Laboratory

- A. Water Quality - Due to real and perceived contamination of surface and ground water supplies by nitrates, areas of needed research are:
1. Increased efficiency of nitrogen fertilizers in order to reduce the amount of nitrates available to be washed into surface waters via eroding sediments or leached below the root zone and possibly into ground water supplies. Includes improved prediction of mineral N requirements of crops in order to reduce the amount of applied N.
  2. Increased information on the form of N being lost with eroding sediments.
  3. Information on the management practices used on various types of soils that would pose a risk of contaminating ground water supplies with agricultural chemicals. A given management scheme may not pose the same risk on different soil types.
  4. Improved methods of soil conservation that will decrease N lost with eroding sediments.
  5. Increase in the active fraction of organic matter in order to increase storage of plant available N. Includes increased efficiency in using N stored in crop residues.
- B. Development of decision support aids (computer models, publications, etc.) to help farm managers in selecting management practices that will strike a balance between profitability and soil and water conservation.
- C. Improved N use by various plant species and cultivars.



**WORKING GROUP 2**  
**FERTILIZER MANAGEMENT**



USDA-ARS Working Conference on Nitrogen Research  
St. Louis, MO  
May 23-25, 1989  
Armand Bauer, Soil Scientist

TECHNOLOGICAL ACCOMPLISHMENTS: Nitrogen Management, Mandan, ND

1. Developed information on N concentration and content of hard red spring wheat (HRS) by plant development stage. The N concentration in leaves and stems decreased and that of viable spike also initially decreased then leveled off at progressive development stages. The N content peaked at about flag leaf elongation and heading stage in leaves and stem, respectively. About 70-75% of total N accumulated in kernels under went a storage period in leaves and stems. This has utility for determining post-emergence N fertilizer scheduling.
2. Developed information on HRS wheat and hard red winter wheat (HRW) water use by plant development stage and vertical downward penetration depth and rate of roots. Water use averaged about 17 cm by onset of heading, or about 50% of total water use to kernel hard stage. Penetration depth, based on measurement of soil water content with a neutron meter, was about 1.52 meters. Vertical downward penetration depth was about 2.5 and 2.3 cm per day by HRS and HRW wheat respectively. When available N has moved deep into soil profile, this information has utility for N fertilizer application scheduling in conjunction with #1.
3. Developed relationships between number of spikes per unit area and grain yield of both HRS and HRW wheat. Number of spikes accounts for about 85 to 90% of variability in grain yields up to about 3000 kg/ha. Estimates of spike number from seedling population has utility in determining yield potential, hence N needs.
4. Developed equivalent information about spring barley as HRS wheat. Spring barley planted on same day as HRS wheat matures about 14 days earlier because it emerges about one day sooner, requires fewer heat units per phyllochron than most HRS wheat, and begins anthesis at about mid-boot stage or about 8 to 9 days earlier in a phenological scale. Utility is the same as described for HRS wheat.

SCIENTIFIC ACCOMPLISHMENT: Nitrogen Management, Mandan, ND

1. Developed information on stress effects during 4 to 5.5 leaf stage of HRS wheat on number of spikelets per spike. Spikelet number decreases with increase in stress. Greatest potential for imposition of stress is air temperature. When combined with #3 under Technological Advancements, this enhances predictive capability to determine yield potential, hence N needs.

USDA-ARS Working Conference on Nitrogen Research

St. Louis, MO

May 23-25, 1989

Armand Bauer, Soil Scientist

Research Activities: Nitrogen Management

The major focus is to evaluate the timing and quantity of fertilizer N application to enhance spring wheat grain yields and protein concentration. Timing fertilizer N application to coincide with plant needs in relation to development stage and available water supplies may improve N use efficiency and simultaneously decrease the potential for ground water enrichment from excess N rates. This study is an outgrowth of research which described N concentration and content in leaves, stems, and spikes by plant development stages from 3-leaf to kernel ripe.

The approach involves field and greenhouse experiments. In the field, the N timing comparisons for yield enhancement are made between a treatment having all fertilizer N to be applied present at planting with treatments having partial amounts present at planting and the remainder applied broadcast, from ammonium nitrate, at two or three pre-heading development stages and at pre-anthesis heading stage. Two or three available N levels, which are the indigenous nitrate-N content to 1.2 m plus fertilizer N, are tested in all combinations with two water levels,

rainfed and rainfed plus supplemental. Treatment effects are measured by N concentration and content in leaves, stems, and spikes from weekly harvests beginning at the 3-leaf stage, and by bi-weekly measurements of N concentration and content in kernels from anthesis to ripe stage. Soil water content is measured in 0.30 increments to 1.8 m, weekly.

For protein enhancement evaluation, two rates of a liquid N source are foliar-applied to all yield-enhancement treatments at the water-ripe stage of kernel formation.

In the greenhouse, comparisons similar to those in the field supplement the information developed in the field.

Southern Plains Range Research Station, Woodward, OK, W. A. Berg

Ave. annual pptn. is 58 cm. Major land uses are grazing of native range, farming to continuous wheat, and grazing warm-season grasses seeded on marginal farmland.

#### Technological Accomplishments

1. Nitrogen applied in April to improved grass pastures was as effective and in some years more effective in forage production and apparent fertilizer N recovery than split April and June application.
2. No-till management of graze-out wheat pasture controlled soil loss within acceptable limits as compared to soil loss of 10 times T under conventional cultivation. Total N lost in sediment and runoff was 7 kg N/ha/yr under no-till and 53 kg N/ha/yr under conventional cultivation over a 2 year period.

#### Scientific Accomplishments

1. Apparent  $N_2$  fixation by alfalfa was 220 kg N/ha/yr over six growing seasons. Sixty percent of the N was harvested in forage and the remainder was residual in crowns, roots, and the surface 10 cm of soil.
2. Cicer milkvetch was marginally adapted to loamy upland soils and not adapted to deep sandy soils. However, apparent  $N_2$  fixation by cicer milkvetch on a loamy soil was high (150 kg N/ha/yr) - this is encouraging for further development of cicer milkvetch for areas where the species is adapted.
3. Soil in N-fertilized improved grass pastures established on marginal farmland accumulated 5 to 10 kg N/ha/year over 20 years. This rate of N accumulation is about one tenth of the estimated rate of N loss under past farming practices. Thus, rebuilding soil N fertility with improved grass pastures is a slow process when compared to the potential for N loss under cultivation.
4. Developed limited information on establishment and management of warm-season legumes for the Southern Plains.

#### Research Underway

1. Effect of N rates on fertilizer N use efficiency in steer gain on improved grass pastures.
2. Effect of urea or ammonium nitrate upon Old World bluestem forage production and forage N accumulation as affected by residue burning, calcareous and non-calcareous soil, and time of N application.
3. Effect of rate and time of N fertilizer application upon production and species composition of native warm-season grass mixtures established on marginal farmland.



### Urgent Problems to be Solved

Problem: Over the past ten years an estimated 4 to 5 million ha of marginal farmland in the Southern Plains has been seeded to grass. Much of this land is deficient in plant-available N after 60 to 100 years of cultivation, cropping, and accelerated erosion.

#### Research Imperatives

1. A viable, long-term research commitment to the selection, breeding, establishment, and grazing management of N<sub>2</sub>-fixing plants for the Southern Plains.
2. Improved N (anhydrous ammonia, urea, N solutions) application methods for pastures.

Problem: Nitrogen and carbon balances for native range and improved pastures need to be calculated to predict long-term effects upon soil fertility from grazing management practices such as early intensive stocking and annual burning.

#### Research Imperatives

1. Quantification of N volatilization losses from urine and feces of ruminants grazing in the Southern Plains.

"Fertilizer Management" research at the USDA Conservation and Production Research Laboratory, Bushland, Texas.

Harold V. Eck

CRIS no. 6209-12000-001-00D "Optimizing soil nutrient status for best water use under dryland, irrigation, and limited irrigation."

Problem: To determine fertilizer needs in conservation tillage systems.

Need for Research: No-tillage, mulch tillage, and other conservation tillage practices are being accepted by producers in the Southern Great Plains. When under conventional or stubble-mulch tillage, Pullman clay loam and other fine-textured soils of the Southern High Plains have been shown to contain sufficient plant nutrients for as high yields as precipitation will allow. However, with increased soil water storage and decreased available N under no-till, yields could be limited by supplies of available N.

Current Research: Conventional tillage (sweep plowing) and no-till are being studied under a wheat-sorghum-fallow sequence, under alternate wheat and fallow, under continuous wheat, and under continuous grain sorghum. The soil N status is being characterized by determining total N and organic matter by 0-7.5, 7.5-15, and 15-30 cm depth increments to 30 cm and  $\text{NO}_3^-$ -N by those increments and by additional 30 cm increments to a 1.8 m depth. Sodium bicarbonate-extractable P is being determined on the 0-7.5, 7.5-15, and 15-30 cm depth increments.

Soil  $\text{NO}_3^-$ -N levels are monitored to a depth of 1.8 m at the beginning and end of the growing season and to a depth of 0.3 m at 3-week intervals during the growing season. In addition, field fertility studies are being conducted on continuous wheat and wheat-sorghum-fallow sequence plots.

Results:

In the three years that the study has been conducted, surface (0-0.3 m)  $\text{NO}_3^-$ -N levels have been similar on no-till and sweep tilled soil while 0-0.9 m and 0-1.8 m  $\text{NO}_3^-$ -N levels have been higher under sweep tillage than under no-till. In the field fertility studies, N and P treatments have not caused significant differences in wheat yields. Vegetative responses have been observed on continuous wheat no-till plots in the spring but limited water has prevented grain yield responses. Average grain yields on continuous no-tilled wheat were 1270 kg/ha in 1986, 2060 kg/ha in 1987, and 1810 kg/ha in 1988. In the wheat-sorghum-fallow sequence, no-tilled wheat yields averaged 1470 kg/ha in 1986, 3660 kg/ha in 1987, and 1540 kg/ha in 1988. Respective average yields on sweep tilled plots were 1380 kg/ha in 1986, 3175 kg/ha in 1987, and 1470 kg/ha in 1988.

Applied N (54 kg/ha as ammonium sulfate) increased yields of both no-tilled and sweep tilled continuous grain sorghum by 57% (from 2645 to 4155 kg/ha). Three consecutive years of above average yields have depleted soil N reserves.

Problem: Phosphorus needs on Pullman soils.

Need for Research: In early fertility studies at Bushland, Pullman clay loam did not respond to phosphorus fertilizer unless the topsoil had been lost. Cropping under irrigation has continued since those earlier studies and some indications of phosphorus response have been observed in recent years. We need to assess the current P status of irrigation Pullman clay loam and determine levels at which P response is obtained and P rates required for optimum yields and water use efficiency.

Current Research: A field experiment is being conducted to determine P needs of irrigated winter wheat on Pullman clay loam.

Results: Hail and insect damage have prevented our obtaining meaningful results during the first 3 years of the study.

### Technological Accomplishments

1. In Fertilizer x Irrigation studies with winter wheat on Pullman soil at Bushland, Texas, N response data showed that 1 kg N (fertilizer + soil N) would be required for each 32.5 kg of anticipated grain yield. This information may be used in adjusting N fertilizer rates to anticipated yield levels when yields are limited by water supplies.
2. Studies with sugarbeets on plots that had or had not received beef feedlot waste (FLW) in previous studies indicated that FLW may have beneficial effects on sugarbeet production on Pullman soil beyond supplying plant nutrients. If so, there would be an incentive to use a material that is now a surplus waste product. This subject merits further study.
3. Alfalfa has been shown to remove  $\text{NO}_3^-$ -N from any depth where soil water is extracted. Since alfalfa frequently roots to depths  $> 6$  m, its use is a management alternative for removing  $\text{NO}_3^-$ -N from the soil at depths below the rooting depth of most crops. Also, utilizing this crop where the  $\text{NO}_3^-$ -N content of the soil is high could reduce the amounts of  $\text{NO}_3^-$ -N leaching from fields to underground water.

## RESEARCH NEEDS

Develop basic strategies for improving fertilizer use efficiency.

- a. Determine the effects of conservation tillage systems on plant nutrient availability and needs.
- b. Determine plant nutrient needs for yield levels attainable under specified climatic and management levels.
- c. Determine most efficient times and methods of applying plant nutrients in conservation tillage systems.
- d. Evaluate use of legumes in cropping sequences to supply part of required N, scrub excess  $\text{NO}_3^-$ -N from the profile, maintain OM and soil tilth, etc.

CURRENT RESEARCH ACTIVITIES:David L. Grunes, Soil Scientist:

Before coming to Ithaca, NY in 1964, I worked for the USDA/ARS in Mandan, ND and for 9 months on a special assignment with the USDA/ARS at Ft. Collins, CO. At the time I was involved in N research on several projects, including the following: the effect of N on the availability of P to plants; the effect of soil moisture on ammonification and nitrification in the soil, and on the uptake of N and P by plants; soil incubation measurements as predictors of N availability to plants; factors affecting N concentrations in field grown plants; comparison of non-legumes and legumes as sources of N for plants; and the efficiency of N uptake by plants from N applied in the field. Since coming to Ithaca, my N research has been concerned primarily with the effect of N source and level on the plant content of minerals important for animal and human nutrition.

Since coming to Ithaca, there have been studies of the effects of source and level of N on the uptake and translocation of Mg, Ca, and K in plants. Some of the grass tetany research has also included studies with grazing cattle. Some of the research has been cooperative with USDA/ARS and experiment station personnel in several parts of the country. There have been growth chamber, greenhouse, and field experiments. Some of the findings are listed below:

Field experiments were carried out, for four years in northeastern Nevada, in cooperation with Henry F. Mayland of the ARS/USDA at Kimberly, ID. Increasing the level of added  $\text{NH}_4\text{NO}_3$  increased Mg and Ca concentrations in crested wheat grass forage. This would help to prevent grass tetany of grazing beef cattle. However, the favorable dietary effects of the higher Mg concentrations may have been offset by increased concentrations of K, N, higher fatty acids, N/total water soluble carbohydrates, and aconitic acid, since these parameters are associated with decreased Mg availability to cattle.

An experiment was carried out with cool season bromegrass at Mandan, ND in cooperation with Ronald F. Follett and James F. Power who were then at Mandan. The  $\text{NH}_4\text{NO}_3$  increased total N, K, aconitic acid, and the ratios of  $\text{K}/(\text{Ca}+\text{Mg})$  and N/total water soluble carbohydrates. Therefore, N fertilization increased the likelihood of grass tetany of grazing beef cattle. In a later field experiment with  $\text{Ca}(\text{NO}_3)_2$  and  $(\text{NH}_2)\text{SO}_4$  as N fertilizer sources, both N sources increased the grass tetany hazard of the forage.

Field studies were carried out with winter wheat forage in Oklahoma and Texas, cooperatively with Bobby A. Stewart of ARS/USDA at Bushland, TX and Floyd P Horn, who was then with ARS/USDA at El Reno, OK. Fertilization with N generally increased the indices that are commonly associated with the high incidence of frothy bloat of male and female younger cattle (stocker cattle), and the wheat

pasture tetany of pregnant and lactating older cattle. Elimination of fertilizer N is not practical because of the pronounced decrease in forage and grain production. Therefore, other management practices are required to minimize or prevent frothy bloat and tetany of beef cattle grazing winter wheat forage.

A greenhouse study was carried out with wheat forage grown in soil, cooperatively with Aubra C. Mathers and Bobby A Stewart of ARS/USDA at Bushland, TX. The nitrification inhibitor Nitrapyrin (N-serve) depressed concentrations of Mg and Ca more than K in the forage, thus increasing the ratios of  $K/(Ca+Mg)$  and causing a forage more likely to cause grass tetany. Nitrapyrin was effective in delaying nitrification, and this could be useful for supplying N to wheat later in the growing season. However, other steps might have to be taken to avoid an increased incidence of grass tetany.

There have been numerous deaths of grazing beef cattle grazing tall fescue fertilized with either high rates of broiler litter, or high rates of N and K fertilizer. A field experiment was carried out with tall fescue at Watkinsville, GA, in cooperation with Stanley R. Wilkinson. The addition of either  $NH_4NO_3$  and KCl, or broiler litter, markedly increased concentrations of N and K in the forage. Fertilization with high rates of N markedly increased concentrations of Mg, and to a lesser extent Ca, in the plants. Fertilization with broiler litter markedly increased the Mg concentrations. Additions of either  $NH_4NO_3$  or broiler litter greatly increased the concentrations of malic acid, and to a lesser extent the citric acid, concentrations. Concentrations of total organic acids were also increased. The high N and K concentrations in tall fescue fertilized with high rates of N and K, or high rates of broiler litter, would make the forage likely to cause grass tetany. The increased organic acid concentrations in these plants might also contribute to some of the grass tetany problems observed when cattle are grazed on fertilized tall fescue forage.

In a growth chamber study at Ithaca, NY, wheat forage was grown with different proportions of  $NO_3$  and  $NH_4$  in the solution culture. It was found that at low Mg levels the Mg concentration in shoots decreased with increasing proportion of  $NO_3$  in the solution culture. Increasing the  $NO_3$  level markedly increased K concentrations in the shoots and roots. Also, increasing the K concentrations in the roots depressed net translocation of Mg from roots to shoots.

In a growth chamber study at Ithaca, NY, leaf lettuce was grown with different proportions of  $NO_3$  and  $NH_4$  in the solution culture. Increasing the proportion of  $NO_3$  in the solution culture markedly increased Ca concentrations in the plants.

As part of a proposed article by ARS/USDA personnel on water quality, I wrote the section on nitrates and nitrites in relation to human and animal health. An extensive search of the medical, animal science, and agronomic literature was carried out to prepare

for this task.

RESEARCH PRIORITIES:

David L. Grunes, Soil Scientist:

Information is needed on interactions of levels of N, K, Mg, and Ca on plant constituents in forages in relation to grass tetany and wheat pasture tetany of pregnant and lactating cattle, and frothy and gaseous bloat of younger cattle. Such studies should include the effects of different sources of N. The additional effects of air and root-zone temperature, and soil moisture level are not yet well understood. Much less is known about the effects of N and other elements, and environmental effects such as air and root-zone temperature and soil-moisture level on the mineral composition of food crops for humans, and that type of research should be carried out.



## Current Research Activities

D. L. Karlen, USDA-ARS  
National Soil Tilth Laboratory  
2150 Pammel Dr., Ames, IA 50011

### I. Assessments of long-term management practices on soil profile N and pesticide distributions.

1. Long-term effects of two different management systems on soil profile  $\text{NO}_3\text{-N}$ ,  $\text{NH}_4\text{-N}$ , and total N concentrations are being determined as a function of soil map unit. The two management systems consist of a 2-year, corn-soybean rotation for which moderate amounts of fertilizer and pesticide are applied, and a low-input system that utilizes a 4-year, corn-soybean-corn-oat/hay crop rotation, manure application, and no pesticides. Soil samples have been collected to an average depth of 4 m across both of these 40-acre sites. Seasonal microclimate, soil fauna, hydrologic gradient, surface and subsurface physical characteristics, infiltration, and crop yield variation are among the parameters being quantified.

2. Soil, plant, and tile drainage water samples are being collected from 36, 1-acre plots near Nashua, IA, to determine nutrient and pesticide loss associated with a continuous corn and a corn-soybean crop rotation. These crop rotations have been grown continuously on these plots since 1977. Data are being collected cooperatively with three ISU scientists in the Agronomy and Agricultural Engineering Departments.

### II. Quantification of the fate of N in a corn-wheat-cotton crop sequence grown on Norfolk loamy sand in the southeastern Coastal Plain.

1. A cooperative project between the National Soil Tilth Laboratory and the Coastal Plains Soil & Water Conservation Research Center is being conducted near Florence, SC, to quantify the fate of fertilizer N applied at various phases of a corn-wheat/cotton crop rotation. The study was initiated in 1988 at a research site that had conventional and conservation treatments established on Norfolk loamy sand since 1979. Replicated  $\text{N}^{15}$  microplots (~7.5 ft. x 9.0 ft.) are being established for both tillage treatments for each crop by applying a 5% label to the fertilizer. Soil and plant samples are being collected, prepared, and analyzed through the ARS laboratory at Lincoln, NE, to determine N uptake from current fertilizer or residual N pools. When fully implemented there will be 48 microplots at this 7-acre research site.

### III. Utilization of mathematical splining to identify amount and timing of maximum N accumulation for various crops.

1. Mathematical splining has been shown to be an effective technique for identifying rates and times at which nutrients are being accumulated by corn (published), wheat (in press), and soybean (in progress). This technique may be useful for optimizing N fertilization timing and rates. It also suggests that N accumulation during the transition from vegetative to reproductive growth either ceases or occurs only in the root system.

## Technological Accomplishments

1. ARS scientists working in cooperation with Dr. Jim Baker at Iowa State University developed a point-injection fertilizer placement device that is effective in crop residues, accommodates split-application throughout the growing season, and reduces the potential for volatilization loss by giving immediate soil contact to the liquid fertilizer. This technology is now being marketed by the CADY Company.
2. Controlled and reversible drainage (CARD) systems in the southeastern Coastal Plain were shown to decrease the  $\text{NO}_3\text{-N}$  concentration of stream water entering a controlled area as compared to that exiting the area, apparently because of increased denitrification.
3. Fertilizer placement through tubes attached to subsoil shanks was shown to be as effective as traditional 2 x 2 fertilizer placement for Coastal Plain soils. This placement technique made planting into crop or weed residues easier because a potential area for entanglement of the vegetation was eliminated from the subsoil-planting implement.

## Scientific Accomplishments

1. Mathematical splining was shown to be an effective method for describing rates and timing of aerial nutrient accumulation by corn. This technique may be useful for improving fertilizer application timing and efficiency of use. It has also identified an apparent period of low aerial N accumulation or possible loss when the plant changes physiologically from vegetative to reproductive growth phases.

## Future N Research Needs for ARS

D. L. Karlen, USDA-ARS  
National Soil Tilth Laboratory  
2150 Pammel Dr., Ames, IA 50011

1. [Problem: Effects of fertilizer management practices on our Nation's groundwater resources are unknown]. ARS and State Experiment Station (SES) scientists have collected a tremendous amount of N rate, form, placement, and time-of-application data throughout the U.S. These research efforts have created an extensive data base with varying climatic conditions. A coordinated program should be initiated to consolidate, summarize, and analyze this information (probably by using selected crop growth or management models). By initiating this type of coordinated effort, the ARS could provide active leadership on the fate of N with regard to our nation's groundwater questions.
2. [Problem: Several aspects of the N-cycle have not been quantified as a function of the soil condition or management practices]. The fate of N during and between growing seasons as a function of soil map unit, tillage system, and soil tilth that can't be quantified by the existing data base needs to be determined with complementary studies in which the same soil, weather, and plant measurements are made at several locations throughout the US.
3. [Problem: The role of cover- and/or catch-crops for the Corn Belt region are unknown]. The effectiveness of cover-crops and/or catch-crops as N sources and sinks needs to be quantified.
4. [Problem: Soil and crop management factors affecting denitrification are not understood]. Improved techniques for assessing denitrification losses in various field conditions need to be developed.
5. [Problem: Recovery of fertilizer N by grain crops is often very low]. Physiological explanations for an apparent lack of aerial-N accumulation during the transition from vegetative to reproductive growth for corn and other crops needs to be quantified. This would help optimize timing and rates of fertilizer application, and thus improve the percentage of fertilizer N recovered by the plant.
6. [Problem: Procedures for assessing residual soil-N status are needed for several types of fertilizer management programs]. Soil-test sampling procedures for assessing residual soil-N status prior to fertilizer application need to be developed for farming systems that are using techniques such as injection of anhydrous ammonia or other band-applied fertilizer sources (injected manure, sludge, etc.).

## FUTURE PROBLEMS AND RESEARCH PRIORITIES FOR ARS

### Fertilizer Management Work Group:

1. Develop basic crop and soil management strategies for improving nitrogen use efficiency.
  - a. Determine the N requirements of alternative crop rotations to wheat-fallow.
  - b. Determine the effects of crop rotations and cropping intensity on water and N use efficiency and economic sustainability.
  - c. Determine the effects of N source, placement, and time of application on crop yield, crop quality, residual soil N distribution and amount, and potential for NO<sub>3</sub> contamination of groundwater.
  - d. Evaluate the effectiveness of applying N fertilizer by soil type for improving crop yields and N use efficiency.
  - e. Improve reliability of N soil test and develop techniques for making site specific N recommendations for all crops.
  - f. Determine N uptake patterns of crop as related to plant growth stage and climatic conditions for improving efficiency of N uptake by crops.
  
2. Develop crop and soil management models for making fertilizer management decisions for optimum crop yield, efficient N use, and economic sustainability.
  - a. Utilize a systems research approach (interdisciplinary) to generate the needed data base from which to develop management decision making models.
  - b. Develop management models that consider the management skills of the farmer for making N fertilizer recommendations that will preserve environmental quality.
  
3. Develop methods for effectively transferring technology to the producer.
  - a. Determine ways to effectively transfer known technologies and intensive management systems (i.e. - maximum economic yield (MEY) concept) to producers.

## NITROGEN CONFERENCE - 1989

G. W. Langdale

Continuous long-term conservation tillage in the humid-eastern U.S. will require new N management strategies. Average annual (double crop) stover production on humid-thermic Ultisols approximates decomposition rate of 10 to 12 Mg ha<sup>-1</sup> yr<sup>-1</sup>. These additions create a gramineous plant response threshold following 3 to 5 years of continuous conservation tillage. Nitrogen in the surface cm depth then increases approximately three-fold (0.05 to 0.15%). Soil carbon increases of this depth are also about 3-fold (0.70 to 2.10%). The soil surface matrix modification also increases rainfall retention to more than 90% on slopes up to 7%. Collectively, new management strategies are essential to support a sustainable agriculture. Crop and tillage relations must be used with holistic approaches to optimize production and sustain environmental quality.

Alan Olness, Morris, MN.

Technological accomplishments:

1. Scientists at Morris, MN developed an isopotential available ion extractor capable of obtaining a charge-weighted mole fraction sample of exchangeable ions under ambient redox conditions.

Scientific accomplishments:

1. Evidence has been obtained that both soil and air thermal energy intensity affect the time and rate of growth and nutrient accumulation by maize.

2. A logistic plant growth model has been modified to incorporate thermal energy intensity. Inclusion of thermal energy intensities transforms a logistic model into a thermal energy driven model of plant nutrient accumulation.

3. Tillage effects soil aeration and soil redox potential below the tilled zone. Non-tilled and ridge-tilled management treatments show better aeration characteristics even with greater than average precipitation.

4. Tillage by hybrid interactions on maize yield have been recorded. Results suggest that hybrid selection should be compatible with tillage-residue management applications.

Soil Nutrient Ratios and Plant Accumulation Abilities

1. Determine effective nutrient availability ratios in soil.
  - a). effect of aeration status on nutrient availability.
  - b). effect of nutrient ratios on nutrient use efficiency.
  - c). effect of nutrient ratios on protein ratios produced within a hybrid/variety.
2. Predict time and intensity of nutrient demands for various crops and varieties (hybrids) within crops using soil and air thermal intensity data.
  - a). Define contribution of soil water content to thermal energy intensity effects on plant growth.
  - b). Define contribution of soil water content to fertility coefficient of nutrient accumulation.
3. Define nitrogen mineralization potentials in terms of physico-chemical properties soils.
  - a). Determine relationship between organic matter content and  $N_0$ .
  - b). Determine relationship between C:N soil ratios and  $N_0$ .
  - c). Define relationship between mineral surface area and  $N_0$ .
4. Determine the effective rate of nitrogen mineralization potential restoration in terms of physico-chemical properties of soils.

USDA-ARS WORKING CONFERENCE ON N, St Louis, MO. May 23-25, 1989

Fertilizer management in conservation tillage systems  
S. C. Rao and Thanh H. Dao, ARS-El Reno, Oklahoma.

### SECTION I: Accomplishments

#### A. Technological accomplishments:

1. Research at El Reno, OK indicated that placement of N fertilizer in narrow bands on the soil surface improved N-uptake by wheat plants grown under no-tillage management. Banding resulted in increased grain protein content and yield.
2. Fall-applied fertilizer N, treated with nitrification inhibitors, enhanced wheat forage growth in the spring, but did not affect grain and straw yield at harvest. The spring forage increase is an added benefit to grazing livestock producers in the Southern Plains.

#### B. Scientific accomplishments:

1. Surface banding of fertilizer N coupled with increased soil moisture in no-tillage improved N uptake and assimilation in wheat leaves during the spring. Increased shoot crude protein content was observed to be mainly due to enhanced leaf nitrate reductase activity. The increased concentration of reduced N has the potential for improving quality and yield by translocating stored vegetative N into the grain.
2. Nitrification inhibitors such as nitrapyrin, applied with urea in the fall improved soil ammonium-N in early spring, minimizing the likelihood of leaching losses. The N accumulation increased wheat forage growth in early spring. However there was no effect on grain quality or yield at harvest.

### SECTION II: Research Imperatives

Problem: Inadequate knowledge and technology for fertilizer placement and of biological processes of the root zone, for improving N-use efficiency in conservation tillage systems.

#### Research Needs or Objectives:

1. Develop improved understanding of the soil microenvironment to enhance N-availability and N-use efficiency with subsurface placement in conservation tillage system.
2. Develop new chemistry, chemical formulations and improved understanding of fate and effectiveness of nitrification inhibitors in major agrosystems.
3. Determine essential N-requirement and uptake periods throughout the crop life cycle and develop high N-use efficiency cultivars for major agronomic crops.



## Fertilizer Management(Gary E. Varvel)

1. Determine fundamental principles influencing fertilizer N use efficiency in cropping systems.
  - a.) Develop techniques to quantify N fertilizer interactions (immobilization) with soil organic matter and crop residues.
  - b.) Integrate N immobilization information with N use by the crop to quantify components of the N cycle in various cropping systems.
  - c.) Utilize information to improve fertilizer N recommendations to maximize N use efficiency and reduce potential for NO<sub>3</sub>-N leaching.

## Current Research Activities (Gary E. Varvel)

Integrated cropping systems research, with emphasis on crop rotation, N management, and cover crop aspects in conventional and conservation tillage systems.

## Technological Accomplishments

- 1.) Scientists at Lincoln determined N fertilization in reduced or no-till winter wheat-fallow cropping systems was not the limiting factor preventing yield responses to the additional store soil water. These results indicated varietal selection was probably of greater importance than N fertilization in reduced tillage systems.
- 2.) Research at Lincoln showed less NO<sub>3</sub>-N leaching potential was obtained in cropping systems including legumes in a rainfed environment. Less NO<sub>3</sub>-N leaching potential can significantly reduce the possibility of groundwater contamination.

## NITROGEN RESEARCH 1989: CURRENT ADVANCES AND FUTURE PRIORITIES

Working Group: Fertilizer Management

### Present N Research Activities:

Scientists: D. T. Westermann\*  
D. C. Kincaid  
T. J. Trout  
C. W. Robbins  
H. F. Mayland  
R. E. Sojka  
B. D. Meek  
G. Kleinkopf (UI)

Location: Kimberly, Idaho

Project:

- A. Summarize and prepare for publication existing data on
  - (1) N fertilization needs of wheat planted in alfalfa stubble
  - (2) N mineralization and scavenging by wheat after annual legumes
  - (3) Review paper on cropping systems to effectively utilize mineralizable N in irrigated cropping systems
  
- B. Nitrogen management techniques for irrigated potatoes
  - (1) Objective 1: Evaluate preplant N fertilization strategies for different irrigation management systems  
Treatments: Irrigation: furrow, sprinkler  
Nitrogen: placement, w/wo nitrification inhibitor  
  
Data: Potato tuber yield and quality; nutrient uptake, petiole  $\text{NO}_3\text{-N}$  concentrations; soil  $\text{NO}_3\text{-N}$  concentrations; water applications; soil temperatures; weather data to estimate ET; water flux below root zone
  - (2) Objective 2: Improve utilization of sprinkler applied N during potato growth
    - a) Evaluate nitrification inhibitor applied with liquid N sources.  
Treatments: N sources, w/wo inhibitor  
  
Data: Nutrient uptake; petiole  $\text{NO}_3\text{-N}$  concentrations; soil  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$  concentrations after application
    - b) Develop simple management techniques to allow growers to accurately predict next N application using soil and/or petiole  $\text{NO}_3\text{-N}$  concentrations that will optimize plant growth rates and yields.  
  
Approach: Develop simple prediction program using existing data; validate and test program by monitoring selected grower's fields.

C. Other projects related to N management practices at Kimberly

- (1) Utilization of N placed in non-irrigated inter-row position.  
*G. A. Lehrs and R. E. Sojka*
- (2) Irrigating to maintain a dry soil layer below the crop's rooting zone.  
*J. L. Wright and B. D. Meek*
- (3) Determine spatial variability of soil  $\text{NO}_3\text{-N}$  during year.  
*G. A. Lehrs*
- (4) Evaluate potato petiole  $\text{NO}_3\text{-N}$  variability in grower's fields.  
*D. T. Westermann and G. A. Lehrs*
- (5) Evaluate laboratory techniques to estimate mineralizable N.  
*H. F. Mayland and D. T. Westermann*
- (6) Determine suitability of existing models to estimate  $\text{NO}_3\text{-N}$  leaching losses under irrigation. *C. W. Robbins, with others*
- (7) Evaluate effects of conservation tillage practices and cropping sequence on N fertilization needs under irrigation.  
*D. L. Carter and B. D. Meek*

## NITROGEN RESEARCH 1989: CURRENT ADVANCES AND FUTURE PRIORITIES

Working Group: Fertilizer Management

### Future Research Needs:

- A. Cropping and management strategies to effectively utilize mineralizable N
- B. Better estimation of N losses during non-cropped period, and management techniques to capture this N
- C. Effect of macro-pores on N leaching losses, especially with furrow irrigation systems
- D. Better "real time" N management techniques
- E. Actual crop needs; total uptake, soil solution N concentrations, effects on crop growth, etc.
- F. Potential for foliar applied N

**WORKING GROUP 3**  
**ORGANIC SOURCES OF NITROGEN**



USDA-ARS Working Conference on Nitrogen Research

St. Louis, MO      May 23-25, 1989

A. L. Black, LD/Soil Scientist

Mandan, ND

Research Activities:

The principal research experiments are conducted in the field on the Area IV SCD Research Farm with plot dimensions sufficient to minimize confounding effects of one plot upon another. The focus of a major portion of the investigations is an ongoing 28 ha experiment (initiated in 1984) involving three replications of: (1) two cropping sequences; spring wheat-fallow and spring wheat-winter wheat-sunflowers (main blocks 147.6 m x 78.7 m); (2) three residue management treatments; conventional tillage, minimum tillage and no-tillage (main plots 49.2 m x 78.7 m); (3) three fertilizer nitrogen levels using  $\text{NH}_4\text{O}_3$  as the source; 0, 22, and 45 kg N/ha for spring wheat-fallow and 33, 67 and 101 kg N/ha for the spring wheat-winter wheat-sunflower rotation (subplots 26.2 m x 49.2 m) and (4) two cultivars; standard check and "best" new cultivar. Tillage treatments are defined in terms of residue maintenance after seeding such that conventional tillage has less than 30% residue cover, minimum tillage has 30 to 60% cover, and no-till has greater than 60% cover. A fourth tillage treatment for the spring wheat-fallow rotation was added in 1987 within the same experimental area to provide a "black" fallow, no-residue (check) treatment for soil erosion and plant disease evaluations.

Measurements include soil water content to 1.5 m at seeding and harvest of each crop, soil NO<sub>3</sub>-N by 0.30 m increments to 3 m depth, NaHCO<sub>3</sub>-soluble soil P (0-0.15 m), plant population at emergence, development stage and plant growth, plant N and P uptake (grain and straw, separately), crop yield and quality factors, weed populations, plant diseases, soil erodibility data near the soil surface and hourly soil and atmospheric environmental data. Detailed observations of root growth, aerial plant development, and soil water dynamics of spring and winter wheat are also being made.

With the assistance of a Research Associate (Soil Scientist), to be hired in 1989, a 5-year data base will be summarized to determine which crop and soil/residue management system uses available water and nitrogen supplies most effectively by limiting N and water movement below the root zone. The goal is to develop efficient nitrogen/water management models for dryland cropping systems.

A second phase of this study is to quantify the long-term changes in physical and chemical soil properties as affected by cropping sequence, tillage/residue management system, and N-fertilization levels over 6-year intervals (1984, 1990, etc.). A soil scientist with expertise in soil microbiology will be added to the research team in FY-90, to conduct in-depth investigations of soil property changes, especially as related to C, N and P cycling.



Re: Material for Working Group Reports

Section I. Accomplishments (Crop Residue Management)

a. Technological Accomplishments

In the last decade, technological advances in tillage and seeding equipment, and in herbicides for weed control have made conservation tillage (minimum-till, no-till) cropping systems (diverse crops in rotation) attainable in the Great Plains. In addition, advances in fertilizer placement/seeding devices provides producers with an array of geometric options for fertilizer applications in relation to the seed in conservation tillage production systems.

These technological advances have made the seeding of winter wheat into standing spring cereal crop residue possible so that winter wheat has become a viable alternative crop within a cropping system for the Northern Great Plains. The standing stubble traps snow, protects winter wheat plants from winterkill, increases soil water supplies and decreases soil erosion from wind and water. (Bauer and Black, 1989 and Black and Bauer, 1989; submitted to Agron. J.)

b. Scientific Accomplishments

1. Management of plant residue on cultivated and grassland-affects soil organic C, N, and P. Bauer et al, (1981, 1987) made comparisons of the effect of stubble mulch- and conventional-till management over the past 25 years and also between grazed and relic (ungrazed) native grasslands on the same soil series. Cultivated land under stubble mulch tillage had C, N and P levels to 45.7 cm soil depth that were 10.7, 9.3, and 8.2% higher, respectively than conventional tillage cropland. Grazed grassland had organic C, N and P levels that were 22.4, 15.8, and 19.4% higher, respectively than conventional tillage cropland. The ungrazed relic grassland had 41% higher C, 3.5% lower N, and 25.6% higher P levels than conventional till cropland. These findings reveal that stubble-mulch farming practices have maintained organic C, N, and P levels and soil productivity at higher levels than conventional tillage systems. The ungrazed grassland had a relatively large buildup of organic C and P, but the marked loss of organic N signals a potential problem that N itself may become the limiting factor in the maintenance of stable organic C in ungrazed or CRP grasslands.

2. Quantity of crop residue returned to the soil in conservation tillage systems has a positive influence on soil chemical properties. As quantity of crop residue increases corresponding increases occurred for soil organic matter, soil nitrogen, available phosphorus, exchangeable potassium, and nitrogen mineralization by soil incubation (Ref: Black 1973, 1979; Ferguson, 1967; and Unger, 1968). These changes along with the associated decrease in soil bulk density, subsequently improved soil fertility and tilth. Thus the quantity of crop residue produced and returned to the soil becomes an important factor in maintaining soil productivity and a sustainable production system.

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USDA-ARS Working Conference on Nitrogen Research

St. Louis, MO May 23-25, 1989

A. L. Black, LD/Soil Scientist

Mandan, ND

Future Problems and Research Priorities:

1. In lieu of evaluating nitrogen-use efficiency (NUE) solely on a one-time individual crop response basis, an exact assessment of the impact of crop rotations on NUE should be made considering all soil and plant N pools. Studies are needed to evaluate the impact of crop rotations on water- and N-use efficiencies, to better quantify the interrelationship of crop root-zone depth and its characteristics to soil water and N content and to quantify the contribution and sources of all N pools to plant N uptake within a defined agroecosystem. Several crop rotation/residue management cycles are needed because of the strong interaction of climatic variability from year to year on C, N, and P cycles.
2. In lieu of characterizing water degradation research problem approaches, attention should be directed toward finding ways to prevent the occurrence, or alleviate, the cause of water quality problems. Researchable examples are: the development of more efficient methods of utilizing crop residues for on site water

conservation and erosion control to conserve organic matter (C, N, P); the development and use of crop rotations that include both shallow- and deep-rooted crops to match water and N supplies; and the timing of application of fertilizer N to coincide with available water supplies and plant need.

3. Research is needed to quantify the long-term changes in soil physical and chemical properties as affected by cropping sequence, tillage/residue management systems, and N-fertilization levels. These types of investigations can only be accomplished in long-term experiments (3 to 6 crop rotation cycles) in order to properly quantify such changes and develop useful predictive capabilities to determine the sustainability and environmental soundness of the agricultural production system being evaluated.
4. Spring wheat producers are finding it difficult to produce and maintain enough surface crop residue to provide adequate soil protection for the 21-month fallow period (particularly the last 9 months). Research is needed to study various short duration cover crop species for use during the growing season of the summer fallow period. There are two approaches; (1) grow a legume or non-leguminous crop from early spring to July, apply herbicide or undercut-till the cover crop and leave overwinter, (2) grow a legume or non-legume crop from August to time of killing frost, leave cover crop present overwinter. For compliance with the '85 Farm Bill, spring oats or barley are currently being recommended and used as a cover crop on summerfallow from August 1 to killing frost. We need

to study water/nitrogen use relationships, soil water recharge overwinter and N availability/cycling for such cover crops and their subsequent effect on spring wheat yields in the Northern Great Plains.

## CURRENT N RESEARCH ACTIVITIES\*

C.E. CLAPP AND R.H. DOWDY,  
SOIL AND WATER MANAGEMENT UNIT, ST. PAUL, MN

Main objective is to investigate the interaction of N fertilization, tillage, and crop residue management on nitrate-N and humic substance transformations and movements within the plant root and upper vadose zones of agricultural soils. Groundwater quality studies were superimposed on existing field plots (initiated in 1980) consisting of N, tillage, and corn residue management comparisons and interactions. Leachate samples are collected by means of porous ceramic cups for analysis of nitrate-N and humic-N. Fertilizer, labeled with N-15, has provided a tracer technique for isolating and separating inorganic and organic components. Humic substances and other N-containing decomposition products of soil organic matter and crop residues are being analyzed and characterized by combinations of biochemical and physicochemical techniques including: elemental and N-15 analyses; UV, FTIR, and NMR spectroscopy; GC/MS analyses; titration curves; and size/shape of macromolecules by ultrafiltration and viscosity measurements. Comparison of observed and calculated results of N transformations and movements in soil and water systems are being carried out using process-oriented simulation models such as NCSWAP and NCSOIL.

A BARD project in cooperation with J.A.E. Molina (University of Minnesota) and two Israeli scientists is in its second year. Experiments are underway to: (1) test whether soil microbial biomass obtains its N from organic-N (direct) or whether assimilation always uses soil inorganic N(MIT); (2) compare extent of turnover of various forms of inorganic N; (3) determine fraction of decomposed soil organic N recycled into microbial biomass (efficiency factor); and (4) determine N turnover during decomposition of plant residues and composted organic wastes in relation to composition and soil type. Experimental results will be compared with simulated data by the direct and MIT versions of the computer model NCSOIL.

### SELECTED PUBLICATION

- Clay, D.E., C.E. Clapp, J.A.E. Molina, and D.R. Linden. 1985. Plant and Soil 84:67-77.
- Clay, D.E., J.A.E. Molina, C.E. Clapp, and D.R. Linden. 1985. Soil Sci. Soc. Am. J. 49:322-325.
- Deans, J.R., J.A.E. Molina, and C.E. Clapp. 1986. Soil Sci. Soc. Am. J. 50:323-326.
- Hadas, A., J.A.E. Molina, S. Feigenbaum, and C.E. Clapp. 1987. Soil Sci. Soc. Am. J. 51:102-106.
- Clay, D.E., C.E. Clapp, D.R. Linden, and J.A.E. Molina. 1989. Soil Sci. 147:(May issue).

\*USDA-ARS Working Conference on N Research, St. Louis, MO, 23-25 May 1989.

## Future Problems and N Research Priorities

C.E. Clapp and R.H. Dowdy,  
Soil and Water Management Research Unit, St. Paul, MN

### I. General:

1. Provide better estimation of N release and availability from crop residues.
2. Measure transformations and movement of soluble organic-N components in agricultural soils.
3. Provide more reliable estimate of  $N_0$  (potentially mineralizable N) for use as best pool of N for N cycling models.

### II. Specific:

1. Determine amounts and turnover of soluble humic substances in soil solution of soils under various tillage and residue management systems.
2. Determine the existence of and subsequently characterize humic/fulvic acid complexes that affect movement of agricultural chemicals in soils.
3. Determine importance of humin fraction of soil organic matter in agricultural soils with respect to pesticide availability and movement.

USDA-ARS WORKING CONFERENCE ON N RESEARCH, St Louis, MO., May 23-25, 1989

ISSUE: Organic N sources: Conservation tillage cereal-legume research  
Thanh H. Dao, S. Rao, W. Lonkerd, R. Meyer, L. Pellack, El Reno, OK

#### BACKGROUND:

A study of residue management practices was conducted since 1983 at El Reno, OK to develop conservation production systems responsive to the needs of cropping-livestock enterprises and to control soil erosion in the Southern Plains. Progress made in the past six years in plant establishment, weed management and N fertilization have enhanced the probability of success of a no-tillage annual cropping system of winter wheat. However, the viability of alternative tillage practices may well depend upon the perception of environmental risks associated with the increased use of agricultural chemicals, the instability of supplies and costs of N fertilizer production. Research at ARS-El Reno has centered upon the issue whether organic sources of N such as crop residues, cover and green manure crops using Leguminosae adapted to semi-arid regions, would allow no-tillage systems to attain sustainability, insuring economic stability of dryland farming and conservation of the land resource base.

#### CURRENT EFFORTS & ACCOMPLISHMENTS:

Best management practices for soil and water conservation and use, residue management and decomposition, N fertilization, and the effects of livestock grazing are being evaluated in three tillage systems. Field plots of wheat-legume associations were established on a Udic and a Udertic Paleustolls under conventional and no-tillage management. Measurements of legumes' fertilizer replacement values, forage and grain production potential in semi-arid areas were made to develop strategies for improving soil properties, alleviating soil physical and biological constraints associated with the combination of livestock grazing and reduction in mechanical tillage.

In annual wheat-legume sequence, N requirements of the wheat crop has partially been met, particularly in the fall. Benefits of the fixed N must cautiously be weighed against the need for water required to establish the next wheat crop in dryland production systems. In mixed cropping-livestock systems, our results revealed a high wheat forage nutritive value and grazed wheat had a higher N requirements than that grown strictly for grain. Conservation studies to improve nutrient flow between soil-plant-animal compartments are being conducted to integrate legumes in a such system.

#### FUTURE RESEARCH:

Problem statement: Inadequate knowledge of environmental and biological processes exists for developing cost-effective technologies for utilizing biologically-fixed N to improve the stability of conservation systems in semi-arid areas.

#### Research needs:

1. Develop water management strategies and crop sequences for incorporating leguminous crops in dryland production of forage-grain wheat.
2. Develop knowledge of soil structure development in cereal-legume associations to alleviate soil physical and biological constraints to crop growth associated with livestock grazing winter wheat-legume systems.
3. Develop knowledge on rhizosphere processes in wheat-legume associations under conservation/no-tillage for optimal N transfer between legume and non-legume crop and complete accounting of the fate of the fixed N.



## SECTION II: Research Imperatives

Problem statement: Inadequate knowledge of environmental and biological processes exists for developing cost-effective technologies for utilizing biologically-fixed N to improve the stability of conservation production systems for conserving natural resources and preventing environmental degradation.

Research needs:

1. Develop water management strategies and crop sequences for incorporating leguminous crops in dryland production of forage-grain wheat.
2. Develop knowledge of soil structure development in cereal-legume associations to alleviate soil physical and biological constraints to crop growth associated with livestock grazing winter wheat-legume systems.
3. Develop knowledge on rhizosphere processes in wheat-legume associations under conservation/no-tillage for optimal N transfer between legume and non-legume crop.
4. Quantify the fate and efficiency of utilization of the fixed N in the companion or subsequent crop, its distribution in N sinks of agronomic importance.
5. Determine the fate of biologically fixed N remaining in the soil-root environment and develop technologies to minimize its effects and reduce its transport in the subsurface environment.

USDA-ARS WORKING CONFERENCE ON N RESEARCH, St Louis, MO., May 23-25, 1989

Organic N sources: crop residues, cover and green manures  
Thanh H. Dao, El Reno, OK

## SECTION I: Accomplishments

### a. Technological accomplishments:

Organic N research and development has been conducted for many years at a number of ARS locations throughout the U.S, including the humid southeast (Auburn-AL, Watkinsville-GA, Mississippi), the Great Plains (Lincoln-NE, St Paul-MN, El Reno-OK), and the Pacific Northwest (Pullman-WA). Leguminous crops were rethought as promising alternatives to commercial fertilizers to insure the sustainability of conservation production systems. Various legumes have been incorporated in major agrosystems either (i) interseeded as a living mulch/companion crop, or (ii) in rotation with varying degree of success across the country and the world. Although interseeding has shown limited effect of fixed N on the non-legume crop, total crop-legume dry matter accumulation provided greater soil protection from erosion than a pure crop stand. In legume-crop sequences, rotational effects and legume N increased subsequent crop yields by soil incorporation of legume residues green manures. However, leguminous cover crops enhanced yields of the subsequent crops under conservation tillage management only in humid regions. In dry, semi-arid climate, biologically fixed N can only partially fulfill the subsequent crop N requirements in Nebraska and Oklahoma studies.

### b. Scientific accomplishments:

Research of interseeded living mulches improved soil erosion control as greater dry matter accumulation in crop-legume associations provided greater surface coverage than the crop alone. However, the biologically fixed N either had limited, no effect and possibly a deleterious effect on the non-legume crop yield under conservation/no-tillage management such as the corn-vetch association.

Scientists have achieved consistent yield increases in summer crops grown in rotation with plowdown winter cover crops and green manures throughout the southeast, the midwest, and the Pacific Northwest. Promising legumes included many genera such as *Trifolium*, *Vicia*, *Pisum*, *Medicago*, *Melilotus*. In the widening-used conservation technology, cover crops also proved very beneficial to no-tillage summer crops in high-rainfall regions.

In annual winter wheat-legume sequence, N requirements of the wheat crop was partially met, particularly in the fall in the central plains of Oklahoma. The benefits of the fixed N must constantly and cautiously be weighed against the need for moisture required to establish the next wheat crop in dryland production systems.

Thanh H. Dao  
El Reno, OK

(2) Organic N sources in cropping-livestock systems:

a. Technological accomplishments:

Organic N research and development of a cereal-legume production system for the dual purpose of forage and grain production has been limited in the US. ARS scientists at El Reno-OK, Clay Center-NE, Billings-MT and St Paul-MN have focused research to develop improved understanding of the flow of nutrients in soil-crop-animal compartments of mixed agricultural enterprises. Nitrogen is the common nutrient involved in the biochemical functions of soil organisms, the plant, and the animal, and thus has been demonstrated as the key to improving overall productivity of integrated cropping-livestock systems.

Much winter wheat pasture research at El Reno of the past decade has resulted in improved understanding of wheat growth response to grazing management and fertilization. Wheat forage has been shown to be highly digestive and exceeds animal maintenance requirements to yield average daily gain of 0.84 kg in cattle. Conservation production practices have also proved very beneficial in stabilizing wheat establishment and insuring dependable forage accumulation in wheat pastures. Grazed wheat has a higher N requirements than wheat grown strictly as a grain crop. Legumes have enormous potential to improve soil N and improve crop growth and thus the availability of forage for animal growth. Limited success was achieved in identifying legumes adapted to our semi-arid conditions. Studies of the stability of mixed purpose winter wheat-legume associations are being evaluated.

b. Scientific accomplishments:

Much winter wheat pasture research at El Reno of the past decade has resulted in improved understanding of wheat growth response to grazing management and fertilization. Wheat forage has been shown to be highly digestive and can yield average daily gain of 0.84 kg in cattle. Grazed wheat has a higher N requirements than wheat grown strictly as a grain crop, and particularly the spring when additional fertilizer N must be topdressed to sustain crop growth and maintain grain yield potential. Legumes have enormous potential to improve soil N and improve crop growth and thus the availability of forage for animal growth. Ley systems is well adapted to Mediterranean-type of climate of Australia and New Zealand. Limited success was achieved in identifying legumes adapted to our semi-arid conditions. Their ability to adapt to no-tillage practices, sustain grazing and survive extreme variability in precipitation and temperature is being evaluated.

USDA-ARS Working Conference on N Research

Wayne Honeycutt  
New England Plant,  
Soil, and Water Lab  
Orono, Maine 04469

Technological Accomplishments:

1. Many fertilizer recommendations now account for the effect of a previous crop or manure addition on the N fertilizer recommendation for a succeeding crop.
2. Various organic sources of N, such as manure and municipal waste, are being used for crop fertilization.

Scientific Accomplishments:

1. The contributions of a previous crop's residues to the N nutrition of a succeeding crop have been determined for some crop species.
2. A mathematically simple approach for predicting N mineralization from organic sources has been introduced and undergone initial field and lab testing. The method, based on heat units (i.e. soil temp.), may provide the grower with knowledge on the availability of N from organic materials, thereby allowing more efficient fertilization practices and reducing the risks of nitrate pollution.
3. Many of the factors affecting N availability from organic sources have been identified. This has provided some predictive capability in describing N mineralization.
4. Techniques for assessing N contributions from crop residues have been developed. This has included the use of  $^{15}\text{N}$  and the N fertilizer replacement value technique.

**Problem:**

1. Determine basic strategies to optimize the cycling and use-efficiency of organic N sources in cropping systems.

**Research Needs:**

a.) Develop and integrate basic knowledge on the combined influence of those factors affecting N mineralization/immobilization turnover from organic sources.

b.) Develop an accurate approach for predicting N mineralization from various organic N sources under field conditions.

c.) Integrate knowledge of N mineralization from organic sources with knowledge of N uptake demands of various crops to capitalize on the synchronization of these parameters.

Research Accomplishments:

1. Nitrogen Accumulation and Bradyrhizobium japonicum Interactions with Soybean and Environmental Conditions

We established that a Bradyrhizobium japonicum strain x soil water status interaction existed in soybean. Strain USDA 110 is considered to be a good nitrogen fixer, but under drought conditions, soybean inoculated with USDA 110 frequently performed worse than soybean nodulated with indigenous strains for nitrogen production and/or yield. This work provided an explanation to variability associated with inoculation, management of water, nitrogen fixation, and yield of soybean in the Southeast. This work has shown that environmental aspects must be considered in the development of any "super strains" of B. japonicum.

2. Crop Residue and Nutrient Management for Soil and Water Systems

We found that determinate soybean grown in the southeastern Coastal Plain nearly always produced 50-90% of its nitrogen from dinitrogen fixation. The percentage was greatly affected by irrigation and/or rainfall, but it was not greatly affected by conservation vs. conventional tillage. We also found that the percentage of total nitrogen removed by seed harvest was often less than 50% and that soybean could annually add from 15-125 kg/ha of net gain in nitrogen to the soil. We proved that determinate soybean was not a low residue producer, particularly under irrigated conditions; generally maximum standing shoot growth was greater than 9 Mg/ha of dry matter. These findings on sandy, low-organic soils were in direct contrast to findings in the midwestern U.S. where soybean is a net consumer of nitrogen because of its low dinitrogen fixation and low vegetative production.

3. Symbiosis and Photobiology

An interactive effect of soybean row orientation and B. japonicum strain on soybean seed yield was established. We found that the spectral composition of light (especially the ratio of far-red relative to red light, which acts through the phytochrome system within the plant) varies with row orientation, and that the soybean shoot-to-root ratio along with root nodulation could be varied by red and far-red light treatments. We proved that the rate of autoregulation of nodulation in the soybean root was altered by the far-red/red ratio in light received by the shoot. We also showed that the sensitivity of the soybean nodulation to phytochrome varied with B. japonicum strain. Row orientation was found to be particularly important for chickpea inoculated with improved Rhizobium strains in the Bihar Province of India. Soil color was also found to be important in the nodulation, seed yield, and net annual balance of nitrogen in soybean and southern pea.

Research Imperatives for Organic Sources of Nitrogen

1. Increase the total amount of surface layer organic matter and the attendant organic N fraction for soils with low organic matter content.
2. Increase the efficiency of nitrogen fixation in forage and grain legumes.
3. Develop cultivars and management practices that allow efficient use of nitrogen in association with irrigation or conservation tillage.

REPORT FOR ORGANIC SOURCES WORKGROUP

FROM ARS-SWC

LINCOLN, NEBRASKA

SECTION I

A. Technological accomplishments.

1. Determined that N fertilizers must be placed below the surface for maximum efficiency with no-till to avoid immobilization in microbially rich surface soil.
2. Found that most of the N in soybean residues became available and was used by the next crop, whereas only a few percent of the N in corn stover became available.
3. Determined that N in hairy vetch cover crop, when incorporated by disking, was mineralized and utilized during the grain-fill period of the next dryland corn crop, eliminating need for N fertilizer.
4. Found that leaving crop residues on the soil surface enhanced the rate of mineralization and uptake of indigenous soil N, probably as a result of maintaining available water in surface soils more days of growing season.

B. Scientific Accomplishments.

1. Found that aerobic microbial processes (nitrification, respiration, mineralization) was maximum when 60% of the soil pore volume is filled with water. At higher water-filled pore space, rates of aerobic processes decline while those for anaerobic processes (denitrification) increase. This seems to hold for all except some tropical (Hawaiian) soils which contain a high percentage of amorphous clay minerals.
2. Found that crop and cropping systems had a major effect on microbial biomass and activity whereas most chemical applications (with exception of a few insecticides) had very little prolonged effects (organic vs conventional farming).
3. Found that microbial biomass and activity was enhanced by leaving more crop residues on the surface and by reducing or eliminating soil disturbance (no-till vs plow). These practices slowed rate of oxidization of all forms of organic matter, thereby increasing soil organic C and N in surface soils.



## SECTION II. RESEARCH IMPERATIVES.

### A. Research Problems.

1. Determine the relative rates of mineralization of N from various organic sources (soil organic N, crop residues, green manures, animal manure, microbial biomass) as an integrated function of soil water, temperature, quantity of organic matter, and C/N ratio of organic matter, and interactions among these sources.
2. From information above, develop computer models capable of predicting amount of N available from all N sources for various soils, climates, and cropping systems.
3. Develop soil management practices that will synchronize the mineralization and availability of N from organic sources with the N uptake requirements of various crops.
4. Develop improved winter legume cultivars that have winter hardiness, earliness of seed set, rapid growth rate at low temperatures, shallow rooted, and other characteristics needed for use as a winter cover crop in the northern half of the United States. *SCS plant materials centers have genetic material that could be used in plant breeding program.*

USDA-ARS & Rodale Research Center  
Jerry Radke & Rhonda Janke

Technological Accomplishments:

1. Research at the Rodale Research Center in eastern Pennsylvania has resulted in the development of several low-input cropping systems. These low-input cropping systems were designed to maximize the use of internal resources produced on the farm (animal manures, legume plow-downs, plant residues, etc.) while minimizing the use of external resources (synthetic fertilizers, pesticides, etc.) which must be purchased. Diverse crop rotations are the basis for these low-input cropping systems. Rotations include legumes to help maintain soil nitrogen and organic matter levels. Rotations also play an important role in the control of weeds, insects, and diseases. Multiple cropping techniques are beneficial in areas with adequate rainfall. Reduced use of synthetic chemicals in conjunction with catch and cover crops should result in reduced ground water and environmental pollution. Economic returns are often higher because of reduced input costs.

Scientific Accomplishments:

1. Researchers have developed cropping systems which maintain yields as high or higher than "conventional" cropping systems by the use of nitrogen fixing legumes in rotation with corn, soybeans, and small grains. Multiple cropping and cover crops provide a replacement for herbicide by out-competing weeds at critical times in the rotation. For example, soybeans can be drilled into a small grain at pre-boot stage resulting in two crops in one season and weed suppression as well.

Research Needed:

1. Nitrogen dynamics and microbial activity associated with the addition of large amounts of organic matter through legume plowdowns, manure applications, and crop residues. The rate of organic matter decomposition and nitrogen availability is crucial to the development of suitable models for use with low-input cropping systems.

2. The development of legumes and other crops specifically for low-input cropping systems. Ideal crop should produce all the nitrogen it needs, use water sparingly, ...etc.

3. Integrated approach to understanding the physical, chemical and biological processes involved in these systems. The soil is a dynamic, living medium and needs to be treated as such. Also needed is a better understanding of soil surface (soil/residue interface) ecology as it relates to cover crop establishment in conservation tillage systems.

4. A better understanding of overwintering and rotational effects. Important processes go on during the winter. Rotational effects over and above nitrogen gains and pest control have been observed but are not understood.

5. The combination of low-input and conservation tillage practices needs to be researched.

6. A better understanding of animal manure properties as they relate to composting as a way to improve the handling of these resources.

7. Nutrient cycling studies/models/budgets at the farm scale level.

Paul E. Rasmussen, USDA-ARS, Columbia Plateau Conservation Research Center,  
P. O. Box 370, Pendleton, OR 97801

PROBLEMS AND RESEARCH PRIORITIES:

Need to determine the long-term effects of crop rotation and organic residues on soil and water quality.

Need a quick accurate soil test for mineralizable nitrogen.

Need to develop functional landscape productivity indexes for determining crop nutrient status and fertilizer need.

Need better methods to predict crop residue, green manure, and rotation effects on nutrient availability and fertilizer need.

Need to reduce yearly variability in production level and N in green manures and legume residues.

Rangeland Soils and Forage Management Research  
High Plains Grasslands Research Station - Cheyenne, WY

A. Current Research Activities Relating to Nitrogen/Organic Matter Dynamics:

1. We are currently involved in a long-term project evaluating the carbon, nitrogen, phosphorus and aggregation dynamics of highly erodible marginal croplands that have been reseeded to grasslands. This study involves evaluating the dynamics of these factors on: (1) newly established grasslands on highly erodible marginal cropland, (2) continuous wheat-fallow marginal cropland, (3) native rangeland, plowed and put into a wheat-fallow cropping system and (4) native rangelands. The design of this study is intended to enable us to evaluate the regeneration of the soil resource of a marginal cropland and the degradation of a newly plowed grassland put into conventional wheat-fallow management. This will give us a better understanding of the role of organic matter in nutrient cycling and its role in aggregation. We are also evaluating the role the various aggregate fractions play in nutrient cycling (as related to organic matter accessibility). This study was established in 1987.
2. We are also evaluating the rate of decomposition of sawmill wastes (sawdust, bark and chips) used as an amendment in the revegetation of bentonite mine spoils. Decomposition of these amendments has been evaluated over a 5 year period and found to be directly related to nitrogen rate, even though primary decomposers are fungi which have been reported to be effective decomposers of high C:N ratio materials because of their nitrogen conservation mechanisms. Spoil moisture content has not had a significant influence on decomposition. We have data on decomposition, mineral nitrogen, total nitrogen and total phosphorus after 1, 2, 3 and 5 years from buried litter bags and intend to have a final set of data after 10 years.
3. We have also been evaluating the effect of long-term low-rate nitrogen (22.4 and 33.6 kg N/ha) fertilization on nitrogen cycling in a native shortgrass prairie site. The long-term fertilization has resulted in increased aboveground vegetation production and increased litter accumulation. It has also resulted in increased nitrogen (kg/ha) associated with the live plant material and litter. The nitrogen fertilization has also resulted in a shift in species composition. However mineralizable-N, microbial carbon and nitrogen and microbial biomass have not shown any response to the nitrogen fertilizer.

B. Future Problems and Research Priorities:

1. Alternative management strategies for CRP lands: 28 million acres of highly erodible marginal cropland have been taken out of production and seeded to grass and/or trees. Management systems need to be evaluated that enable the landowner economic benefits from these lands without plowing them out and returning them to conventional crop management systems. Benefits in the soil resource base that have occurred during the CRP contract period must be protected to ensure that utilization of these lands does not result in their continued degradation. These studies need to evaluate the need for nitrogen fertilization, interseeding of legumes, alternative cropping strategies and non-traditional use of lands.
2. Better basic data and knowledge of the interactions between soil, plant, water and animal in relation to nutrient cycling and the effects of management alternatives on the soil-plant-water-animal system. Especially important in the management of lands where forages are utilized through grazing.

Technological Accomplishments -- Organic Wastes

1. Scientists at Bushland, TX found the advance of irrigation water in graded furrows was slower and water intake greater on manure-treated soil. Increased grain sorghum yields on manure-treated fields were attributed to both increased soil fertility and soil physical properties.
2. Application of feedlot manure to a clay loam soil at Bushland, TX increased soil organic matter and hydraulic conductivity, and decreased bulk density. Yields of irrigation sorghum, corn silage, and wheat showed that annual applications of 22 t/ha were adequate for maximum yields. Incorporation of the manure immediately after spreading was essential for minimizing gaseous nitrogen losses and preventing degradation of runoff water quality.

Research Accomplishments - Organic Sources

1. Poultry litter applied at rates greater than 4 tons per acre to tall fescue pastures results in increased incidence of animal health problems such as grass tetany, and nitrate toxicity to ruminant animals. (Maximum loading rate to prevent animal health problems on cool season forages.)
2. Poultry litter applied to Coastal bermudagrass hay fields at rates of 10 tons/acre over a 4 year period result in excessive accumulation of  $\text{NO}_3\text{-N}$  in soil solution below the effective rooting depth of Coastal bermudagrass.
3. No-till planting of rye (cereal grain) in the fall in Coastal bermudagrass fields receiving heavy applications of broiler litter significantly reduces losses of  $\text{NO}_3\text{-N}$  to percolating water by scavenging available N left over from the summer crop season.
4. There is less available N for crop uptake, or percolation to ground water in broiler litter than  $\text{NH}_4\text{NO}_3$  for the same level of N inputs. The reduced immediate availability means a higher residual value, and a lower chance of large losses of  $\text{NO}_3\text{-N}$  to ground water.
5. Broiler litter and  $\text{NH}_4\text{NO}_3$  fertilization at 1344 kg N/ha/yr for 2 yrs resulted in similar N recovery efficiency when both were cropped in residual for five years (61% vs 59%).
6. Application of N at 224 kg N/ha/yr did not affect soil  $\text{NO}_3\text{N}$  below the effective rooting depth of Coastal bermudagrass, when N was increased to 448 kg N/ha/yr, the  $\text{NO}_3\text{-N}$  concentration at 122 cm gradually increased thereby showing that the pool of mobile N was increasing, and that after 2 or more years this rate of N would result in  $\text{NO}_3\text{N}$  accumulation greater than 10 ppm  $\text{NO}_3\text{-N}$  below the effective rooting depth of bermudagrass.
7. Organic wastes have been characterized, and variability has been documented. For precise applications, and critical environmental impact statements the amount of N in the waste must be determined.
8. Up to 60% of the N contained in organic wastes may be lost by volatilization as  $\text{NH}_3$ . Amount lost is affected by (1) pH of the organic waste and soil, (2) soil conditions, (texture, permeability, water content), and (3) climate.
9. The concept of a decay series to describe the quantities of N mineralization from an organic source (a first approximation).
10. Holistic approaches to problem solving waste management systems involving whole farms have been developed.
11. Winter legumes can contribute up to 100 kg N ha<sup>-1</sup> to a summer animal grain crop. Nitrogen recovery in a single crop season varies (up to 83%), and legume green manure crops can supply from 40% to 100% of grain crop N.
12. Unaccounted for N losses are greater for organic N sources than for fertilizer N sources.

13. As a first approximation, about 1000kg organic residue per ha is required to prevent soil erosion by water, and this amount increases with increasing slope. Organic N sources are useful for this purpose, and on soils without working N capital (soil organic matter) they are more valuable than their nutrient content.
14. Ability of winter legumes in supplying available N depends on the C/N ratio in the residue, and cultural practices to assure adequate stands of N - fixing (inoculated) plants.
15. Organic sources of N are especially effective in improving soil productivity in soils having marginal, or poor physical qualities for crop production.
16. Loading rates for application of organic wastes to land must be based on chemical analysis, and purpose of waste application and site or location. For instance crop production limits in Oklahoma, or Texas may be determined by salt content, in Georgia by N content, or in the case of sludge the presence of industrial or domestic impurities. Regulations and guidelines are being developed that are very realistic (see proposed EPA Regulations).
17. Organic wastes have been fairly well identified as to probable chances for pathogen contamination, heavy metals, and loading rates or hazards associated with each kind of waste.
18. Systems of waste management have been designed to control pollution from animal wastes, and to prescribe environmentally safe land application rates of sewage sludge (proposed EPA Regulation).



Research Imperatives - Organic Sources

1. Quantitative assessment of Nitrogen economies in grazed pastures, and significance of N return in animal and plant residues.
2. What is the critical organic matter level for soils to provide the working N capital to sustain productivity. What role can organic sources of N play in renovating and restoring marginal soils to their potential productivity?
3. Define the role of organic sources of N in best soil-, water-, and crop management for land with (a) no natural, or indigenous limitations to crop production, (b) few, but significant limitations to crop productivity, and (c) severe, and/or many limitations to crop productivity. Includes coordinated regional research which defines site and climatic resource potentials, and systematic management system development for the more important situations from productivity and environmental quality viewpoints.
4. Cultural and management practices to assure consistency in excellent legume stand establishment, and N fixation there of (productivity as well).
5. Amount of N transfer between legumes and grasses in mixed cultures, and management practices which optimize the N transfer.
6. Methods for techniques to assess N availability from organic sources (improvement of N decay series, or refinement with realistic field verification).
7. As a research organization, we don't set policy, but we do need to provide useful information in priority setting, and attitude adjustment with regard to the use of wastes and water. Each generation must take care of its wastes, but it is unfair for future generations to have to deal with our wastes, or consequences of our poor waste management. Sustainability, and its costs and where and how organic sources can be valuable resources needs further documentation, and explanation. The use of large quantities of water to remove our wastes needs further examination, and exploration of alternatives.



**WORKING GROUP 4**

**WATER QUALITY**



## CURRENT NITROGEN RESEARCH ACTIVITIES AND FUTURE PLANS

David L. Carter  
Soil and Water Management Research Unit  
Kimberly, Idaho

### 1989 RESEARCH

I. Measure nitrate concentration in the soil, nitrogen uptake by sweet corn, and estimate nitrate leaching on the following treatments, for the 1989 growing season.

- A. Alfalfa sprayed and plowed under in the spring, 1989
- B. Same as A, but with 100 lbs N added per acre
- C. Alfalfa sprayed and corn planted without tillage
- D. Same as C, but with 100 lbs N added per acre

For comparison, the following measurements will be made.

- E. Nitrate concentration in the soil throughout the growing season on plots treated as in A and C above, but without a crop.
- F. Nitrate concentration in the soil throughout the season on plots with growing alfalfa. Nitrogen uptake by the alfalfa will not be measured.

II. Measure nitrate concentration and water flows in drainage tunnel and tile effluents for a large irrigation tract and compare with results obtained during the period 1969-1971.

- A. Nitrogen outflow from the tract will be determined
- B. Nitrogen application rates will be estimated
- C. Changes in fertilizer nitrogen application over 20 years will be estimated.
- D. The impact of nitrogen fertilizer use rate change, if any, on the nitrate outflow in subsurface drainage water will be determined.

### FUTURE RESEARCH PLANS

I. Cropping sequences and tillage systems will be developed to enhance the utilization of symbiotically fixed nitrogen by growing crops and reduce nitrate leaching out of the root zone.

- A. Timing the killing of legumes so that nitrogen will be mineralized to nitrate at the right time for most efficient utilization by the growing crop will be one approach to be investigated.
- B. The effect of the method used to kill the legumes on the time that the nitrogen becomes available to the growing crop will be studied.

II. The possibility of chemically controlling the conversion rate from symbiotically fixed nitrogen to nitrate in the soil solution will be investigated.

# IMPORTANCE OF GASEOUS N REACTIONS AND TRANSPORT IN SOIL-PLANT SYSTEMS ON N-USE EFFICIENCY

Soil-Plant Nutrient Research Unit  
Fort Collins, CO

Wayne Guenzi

The basic research is concerned with understanding the phenomena, principles, and processes that control soil N transformations. Objectives are to identify and quantitatively characterize interrelationships between soil and environmental factors on the production or consumption of various nitrogen intermediates involved in nitrification and denitrification processes. The approaches are: control of redox potentials at specific levels at constant pH and temperature, and evaluation of NO and N<sub>2</sub>O production/consumption as a function of inorganic N-oxides; evaluation of the effects of low O<sub>2</sub> concentrations on the kinetics of gaseous N production/consumption in relation to NO<sub>3</sub><sup>-</sup> and NO<sub>2</sub><sup>-</sup>; and inclusion of pH and temperature as variables while controlling other factors. Laboratory studies are conducted in a specially designed apparatus that automatically maintains redox, pH, and temperature at any desired level in a continuously stirred soil slurry. Concentrations of gases are determined by gas chromatography (O<sub>2</sub>, CO<sub>2</sub>, N<sub>2</sub>O, and N<sub>2</sub>) and by a chemiluminescent analyzer (NO and NO<sub>2</sub>). When applicable, <sup>15</sup>N-labeled compounds are used and subsequent analyses made by mass spectrometry. In collaborative laboratory studies, evaluate the contributions of chemoautotrophic ammonium-oxidizing, nitrite-oxidizing, and heterotrophic nitrifying bacteria on NO and N<sub>2</sub>O emissions from aerobic soils. Field studies will include studies on gaseous emissions of NO and N<sub>2</sub>O, and factors controlling soil NO<sub>3</sub><sup>-</sup> distribution in a corn field irrigated with a center pivot adapted with a low energy precision application (LEPA) delivery system.

#### SOIL NITROGEN RESEARCH NEEDS:

1. More effort needs to be placed on denitrification losses. This effort should focus on both basic research identifying factors controlling type and amount of gaseous N products and field studies to evaluate amounts of gaseous N products ( $\text{NO}_x$ ,  $\text{N}_2\text{O}$ , and  $\text{N}_2$ ) from highly fertilized crops. Having a good idea of expected gaseous N losses would be helpful when assessing fertilizer needs for a cropping season, and would also provide much needed information on agriculture's role in adding  $\text{NO}_x$  and  $\text{N}_2\text{O}$  to the atmosphere.
2. Little is known about nitrogen transformations below the root zone. With increased emphasis on nitrate pollution in ground water, research is needed on N transport and other factors that may be responsible for nitrate reduction to gaseous products (chemically or microbially).
3. A good, quick, inexpensive method needs to be developed to evaluate the mineralization potential of a soil. This would be a valuable bit of information for a farmer planning his fertilizer N requirements. Not only would this be important from an economic point of view, but would have a positive effect in avoiding over fertilization and thus minimizing nitrate movement below the root zone.
4. Cooperation of agricultural engineers, plant nutritionists, and soil scientists could make an important contribution in optimizing water management and N fertilization with center-pivot irrigation systems. With automated irrigation scheduling along with timely N inputs, it would seem that a high N use efficiency could be obtained.

## CURRENT RESEARCH ACTIVITIES

Richard Lowrance  
Ecologist  
Southeast Watershed  
Research Laboratory  
USDA-ARS  
Tifton, GA

My current research activities are carried out in two hydrologically distinct subregions of the Gulf-Atlantic Coastal Plain in Georgia: the Fall Line Red Hills and the Tifton Upland. The Fall Line Red Hills subregion includes extensive agricultural soils which provide recharge to the Claiborne Aquifer, a major water supply source in the upper coastal plain. Four related projects in the Fall Line Red Hills are designed to provide information about the fate of nitrate from agricultural management in the recharge zone. The projects in this area are: 1) nitrate leaching under simulated rainfall from three tillage systems; 2) nitrate movement to groundwater from an irrigated corn watershed; 3) factors controlling denitrification in soils, subsoils, vadose zones, and aquifers; and 4) development of a monitoring technique for nitrate leaching using anion exchange resins. The Tifton Upland is an important agricultural area which extends in a broad band across the upper coastal plain of Georgia and into South Carolina. The Floridian Aquifer, a major limestone aquifer with perhaps the largest supply of potable ground water in North America, is confined beneath the Tifton Upland. Research activities in this subregion include: 1) processes controlling denitrification and nitrogen removal in riparian forest ecosystems; and 2) comparison of nutrient loads from mixed cover watersheds with different land uses.



## FUTURE RESEARCH PRIORITIES

Richard Lowrance  
Ecologist  
Southeast Watershed  
Research Laboratory  
USDA-ARS  
Tifton, GA

My perspective on future nitrogen cycling research is based on my experience in water quality and watershed research. Based on this background and the nationwide concern about adverse water quality effects due to agriculture, I feel strongly that future nitrogen management research should clearly emphasize nitrate leaching and should be integrated with other environmental quality investigations. There are at least three lines of research that I would like to help pursue in the future:

- 1) Low input sustainable agriculture (LISA) can be divided into two general management approaches - alternative chemical management and alternative farming systems. Shifts to either approach can potentially affect the quality of either surface or ground water in agricultural watersheds. LISA approaches will not necessarily have beneficial effects on water quality unless water quality problems are targeted and management systems developed to address them. I would like to see ARS provide the leadership in examining the water quality and general environmental quality effects of LISA systems.

- 2) Water quality problems in agricultural watersheds are not generally caused by just one field or farm, but are caused by the aggregate land use and the spatial distribution of sources and sinks of agricultural chemicals and nutrients. Theoretically, it

Nitrogen Research Needs

A. Good groundwater sampling techniques

The most difficult step in doing nitrogen studies in groundwater is taking a sample and knowing what that water sample represents. For example, samples can be obtained with ceramic suction cups, but the samples are probably water from the soil matrix. The size of pores from which water is extracted depends on the tension applied, and it's difficult to know whether the matrix water is moving down below the root zone. The relevance of the water sample to groundwater is difficult to assess.

B. Knowledge to relate nitrogen in water to its source.

Being able to measure  $\text{NO}_3$  in water does not indicate whether that N came from fertilizer, animal waste, or mineralization of organic matter.

C. Knowledge to relate soil N (and various forms of soil N) to:

1. Groundwater Quality
2. Plant uptake
3. N fertilizer recommendations (an N soil test)

## Current Nitrogen-Related Research at Coshocton

### A. Pasture Research

A major portion of our nitrogen research involves pastures. Beginning in the early 1970's, we had an area with four watersheds which received 50 lbs. of N/Ac each year, had cattle rotated through all four in the summer, and had the cattle wintered on just one of the watersheds - a five-year study. For the past ten years, three of the four watersheds in this same area have received 150 lbs. of N/Ac each year. The N was broadcast in three applications on the summer only grazed watersheds. Two of the watersheds received methylene urea, a slow release fertilizer furnished by O. M. Scott & Sons, while one watershed received  $\text{NH}_4\text{NO}_3$ . The summer grazing and winter feeding watershed received no mineral N fertilizer. It's N came from the hay that was brought in to feed the cattle. Water samples from surface runoff are analyzed for  $\text{NH}_4\text{-N}$ ,  $\text{NO}_3\text{-N}$ , and Org-N; and water samples from developed springs are analyzed<sup>3</sup> for  $\text{NO}_3\text{-N}$ .

Another pasture study which was begun in the 1970's involved rotating cattle through four fields of orchardgrass (contained two watersheds with developed springs) during the summer and through four fields of tall fescue (contained two watersheds with developed springs) during the winter. During the winter, the cattle ate Fall regrowth and the hay which had been produced on that watershed during the summer. During this five-year study, 200 lbs. of N/Ac were annually applied, split in three applications. The  $\text{NO}_3\text{-N}$  levels in the springs were unacceptably high, and so alfalfa was interseeded in the orchardgrass and tall fescue to provide N. Phosphorus and potassium were added according to soil tests to maintain nutrient balance. As with the above pasture study, surface runoff and water from developed springs are analyzed for nitrogen. Last year (1988) was the ninth year for this treatment.

### B. Conservation Tillage Research

Currently, we have a conservation tillage study involving six watersheds; two are paraplowed; two are chisel plowed; and two are in no-till. This year (1989) will be the sixth year in this corn-soybean rotation; 1989 will be a soybean year. Surface runoff and erosion are measured and analyzed. Seven lysimeters receive the same treatment as the chisel plowed watersheds; percolate is collected and analyzed for nitrogen.

### C. Macropore Research

Undisturbed soil columns were taken at two depths, 35 cm and 70 cm, from a long-term no-till area and a rotationally cropped area. The columns were fitted with percolate collection facilities and replaced in the soil. Nitrogen fertilizer and tracers were added with surface treatments simulating no-till and conventional tillage. Percolate, which resulted from natural rainfall, is being collected and analyzed.

Undisturbed cubes (30 cm on a side) are taken from a long-term no-till area, have the cube bottoms "mapped" for macropores, and are placed on a collection grid with 64 squares. Known amounts of water are added at specific rates by means of a specially design rainfall apparatus. Percolate is collected from each of the producing grid squares and can be related to the "macropore map." For each square, the total amount of percolate and the time from the "rainfall" until the start of the percolation are recorded. Nitrogen fertilizers and tracers are being used in connection with these water flow studies and with different sequences/time intervals of small and large rainfall events. The percolates are available for analysis.

## **Jim Schepers**

### Technological Accomplishments

1. Research at Lincoln revealed that half or less of the nitrate contained in ground water used for irrigation of corn was utilized by the immediate crop. In contrast, the apparent crop recovery of the water borne nitrate ranged from 90 to 60% for the low and high N fertilizer regimes, respectively. Reasons for the lower than expected tagged-N recovery are attributed to immobilization of a portion of the applied nitrate and to the fact that irrigation water was not required until later in the season when the crop had already taken up ~60% of the total for the season. This information is useful to producers and consultants as they strive to improve N management practices by developing better synchronization between N availability and crop N requirements.

### Scientific Accomplishments:

1. Scientist have been able to gain a more complete appreciation for N mineralization by comparing laboratory procedures used in the U.S. with the electro-ultrafiltration procedure used in West Germany and Austria. Nitrogen extracted by laboratory procedures contains both microbial biomass N and N cleaved from organic compounds. Therefore time of soil sample collection and climatic conditions can affect the amount of N recovered in the potentially mineralizable N (PMN) determination. Estimates of PMN represent a point in time while mineralization is an on-going process throughout the year, therefore N management strategies must strive to integrate climatic adjustments into PMN data to estimate crop N availability from mineralization.

Problem: Excessive fertilizer N application because of inadequate procedures to evaluate crop N status in time to correct N deficiencies.

1. Develop easy, rapid, and reliable procedures to sample and analyze corn plants for a good N status indicator.
2. Evaluate the effect of stage of growth, plant part, and cultivar on the value and potential application of tissue testing for N status in corn.
3. Develop sampling and tissue testing strategies that integrate site specific factors (i.e. soil, water, temperature, stage of growth, plant part, cultivar, etc.) into the recommendation.

Current Research Activities:

R. Schnabel, NWRC, Univ. Pk., PA

Following is a list of nitrogen—related research topics and specific CRIS objectives at the Northeast Watershed Research Center

- N use and losses in soils of differing drainage classes

Quantify the effect of soil drainage class on fertilizer N use efficiency and leaching potential, with special emphasis on denitrification and N immobilization.
- GIS and probability kriging to identify and delineate critical areas

Estimate spatial and temporal variability of the major soil and landscape properties and model parameters affecting percolate, NO<sub>3</sub> leaching, and crop yields using Geographic information systems (GIS) and remotely sensed data.
- Groundwater model to assess impact of altered land use practices on water quality.

Develop a basic watershed—scale model of subsurface flow and chemical transport processes tying source, sink, and dilution zones to impact zones (i.e., groundwater wells, streams) through typical Northeast watersheds.
- Aquifer properties and morphology relative to watershed response to inputs

Develop tools to characterize and predict flow and chemical transport through fracture zones.
- Use of natural tracers to determine source areas and travel rates of contaminants

Test the use of natural tracer techniques for analyzing flow sources and pathways and their use to validate the model framework.
- Development of methodology to measure percolate quantity and quality

Quantify percolate from soils and the relationship between percolate quantity and macroporosity.

Quantify NO<sub>3</sub> transport through intact soils containing preferential and rapid flow—through zones
- Denitrification in riparian zones

Quantify the scavenging capacity of shallow ground water and riparian zones , with special emphasis on denitrification and N immobilization.

## Future research problems:

R. Schnabel, NWRC, Univ. Pk., PA

- Develop crop management systems which minimize residual soil nitrogen

Research in the Eastern US demonstrates that a large majority of groundwater recharge and nitrogen export from agricultural watersheds occurs during winter and early spring. Minimizing the concentration of nitrate in solution during this time period, either through synchronizing N applications with N use or by immobilizing residual N in overwinter crops, would favorably impact N loss from crop lands.

- Develop information systems which merge soil, geology, hydrology and chemical information to delineate areas requiring careful management.

Combinations of soil properties, geology and hydrology define the susceptibility of areas to groundwater contamination. Identification of these locations is important to the siting of wells, feedlots, etc.

- Determine the mechanism of groundwater contamination in limestone areas. Percolation through the soil body or entry through sinkholes?

Contamination of limestone aquifers by agriculture is receiving increasing press attention. Sinkholes are thought to provide easy access of surface contaminants to groundwater. If this is the mechanism for groundwater contamination, then management practices or structures must be developed to buffer sinkholes from Ag lands.

- Develop methods to maximize the clean up of waters possible in riparian zones and reduced zones in groundwater.

Processes at off-field locations of a watershed can reduce the nitrate concentration of waters leaving the field. Given that N loss is likely to continue even from well managed lands, options for utilizing and enhancing the clean up of groundwater at off-field locations should be explored.



**WORKING GROUP 5**

**LOSSES OF NITROGEN TO THE ATMOSPHERE**



May 20, 1989

Subject: USDA-ARS Nitrogen Working Conference

To: Gary Heichel

In response to your April 26, 1989 letter, I am enclosing the following short statements.

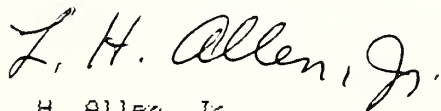
RESEARCH PROGRAM AND INTERESTS. My research program will be directed toward assessing the impacts of Global Climate Change. More specifically, assessing the impacts of increasing temperature, increasing carbon dioxide concentration, and changing water availability on crop physiological responses and agricultural productivity. In our work on climate change effects on rice grown in controlled-environment chambers (SPAR units), we are working with the University of Florida soil scientists in evaluating the losses of nitrogen by soil, nitrogen uptake by plants, and fertilizer N use efficiency, using N-15 techniques. We may also expand later to evaluate effluxes of methane, since methane is rising in concentration and has been identified as a greenhouse effect gas. One prominent source of methane appears to be paddy rice culture.

FUTURE N RELATED RESEARCH THAT ARS SHOULD DO. I will focus my comments on aspects of global climate change and atmospheric trace gas cycling, which is mainly in the area of Working Group 5, Losses of Nitrogen to the Atmosphere.

Since nitrous oxide (N<sub>2</sub>O) is a greenhouse effect gas, and since N<sub>2</sub>O of the atmosphere is gradually rising, ARS could conduct studies of the flux of this gas to the atmosphere, along with fluxes of other nitrogen compounds, under a range of agricultural conditions. This really should be a component of nitrogen cycle studies (including deposition/uptake) rather than a stand-alone project. Perhaps the attention should be on expanded objectives and rationales, rather than on a new program without tie-backs to integrated program objectives and rationales in nitrogen cycle research.

I hope you have a successful workshop. I am sorry that I cannot attend. Pardon my brief comments.

Sincerely yours,



L. H. Allen, Jr.

cc: T. R. Sinclair  
W. Heck  
R. Follett  
D. Meister  
J. Power

USDA-ARS Working Conference on Nitrogen Research  
Working Group on "Losses of N to the Atmosphere"  
**Current Research** - John W. Doran  
May 1989

Current research efforts center mainly on the effects of tillage, cropping, and residue management on the soil physical environment and its influence upon microbial denitrification.

Specific efforts are directed towards reduced-input management systems which utilize legume and grass cover crops to fix N and enhance uptake of N when the grain crop is not growing. Zonal tillage management (Ridge tillage) is used to control weeds and accelerate mineralization of organic N during the growing season.

Past research efforts have indicated that the integrated effects of soil water content and soil density are an important determinant of the potential for denitrification losses in the field. Current measurements of Denitrification using intact soil cores and acetylene blockage indicate that only when the proportion of soil pore space which is water-filled exceeds 70% does significant denitrification occur. With ridge-tillage management this condition exists mainly in the between row area and especially in the wheel track.

Current measurements of denitrification are being related to the soil physical environment as related to time of season, depth in soil, and horizontal variations in tillage and residue management.

## USDA-ARS Working Conference on N Research

### Future Problems and Research Priorities

#### I. Field research to understand the dynamics of gaseous N losses

NH<sub>3</sub> loss and denitrification are dynamic processes that depend upon chemical, biochemical, and environmental factors that are difficult to reproduce in the laboratory, and should therefore be addressed with field-scale research

#### II. Validate field measurement methodology

A number of field measurement techniques have been used to measure N losses (chambers, micrometeorological flux estimates, small plot - horizontal flux or simulation models). Need to verify when the various methods are appropriate, and where the pitfalls are.

#### III. Model N losses

Need a long-term research thrust to understand and begin to model the complex and dynamic processes leading to gaseous N losses

#### IV. Wet and dry deposition of N to lakes and estuaries

To understand the relative importance of agricultural sources of N to lakes and estuaries, such as Chesapeake Bay, numbers for wet and dry deposition are needed. The washout and direct uptake of NH<sub>3</sub>, NO<sub>3</sub>, and other forms of N from the atmosphere may be substantial.

Dwight E. Glotfelty

Environmental Chemistry Laboratory, NRI

Beltsville, MD

Dwight E. Glotfelty

Research Chemist

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Current Research:

Post-application losses of pesticides to the atmosphere

Determining the physicochemical and environmental factors controlling pesticide volatilization

Field measurements of losses

Evaluation of small plot techniques for measuring pesticide volatilization

Developing "eddy accumulation" method for measuring pesticide flux, and comparing results to standard aerodynamic profile flux measurements

Atmospheric transport and deposition

Factors controlling atmospheric transport, residence time, and redeposition of pesticides in the environment

Distribution of pesticides and other organic xenobiotics in fog and clouds

measurement of washout coefficients for pesticides in precipitation

physical chemistry of dilute aqueous solutions of pesticides, including precise measurement of air/water distribution coefficients (Henry's Law Constants)

Aerial deposition of illegal pesticide residues to non-target crops

Atmospheric loading of pesticides and other organic xenobiotics to Chesapeake Bay

Modeling pesticide losses and atmospheric transport and deposition

NITROGEN CONFERENCE 1989

Lowry Harper, Watkinsville, GA 30677

I. Research Interests:

1. Study of N dynamics of Agricultural ecosystem using systems analysis
  - A. He will emphasize the atmosphere component in the system's analysis. Interest in  $N_2O$ , aerosols containing N, and non-gaseous particals.
  - B. He will be working until late fall in the Netherlands.

II. Research Priorities

1. Quantitative description of the atmospheric component of the N cycle.
2. Potential relationships between the N cycle, and the carbon cycle.

Working Conference of Nitrogen Research  
Walter W. Heck  
(Air Quality Research Unit)

1. Statement of Current Research Activities

Conduct research on the effects of atmospheric chemicals (anthropogenic and natural) on agricultural productivity and environmental quality throughout the U.S. These include the effects of airborne nutrient, toxic, acidic, climate altering, and growth altering chemicals on the visible conditions, health, productivity, genetic diversity, species composition, and geographic or economic distribution of agricultural crops. Current primary concerns are ozone, acidic deposition, CO<sub>2</sub> and UV-B irradiation; gases and aerosols from agricultural and other operations; chemicals released into the atmosphere from vegetation, animals and soils; volatilization or incineration of domestic, industrial or municipal wastes; and, chemicals and other changes associated with climate modification.

2. List of Future Problems and Research Priorities

This briefly shows our research interests relating to N compounds that are found in the atmosphere. The compounds of primary concern are: NO<sub>x</sub>, N<sub>2</sub>O, HNO<sub>3</sub> vapor, NO<sub>3</sub><sup>-</sup> and NO<sub>2</sub><sup>-</sup> aerosols, NH<sub>3</sub>, NH<sub>4</sub><sup>+</sup> aerosols, and various N containing organic compounds. Our primary research interests are effects on agricultural systems, sources - especially as related to agricultural activities, and atmospheric reactions.



## Future Problems and Research Priorities for ARS

G. L. Hutchinson

1. Because (1) gaseous N oxides are involved in "greenhouse warming" as well as atmospheric ozone production/destruction, (2) microbial processes in soil is one of the principal sources of atmospheric N oxides, (3) emission of NO (rather than N<sub>2</sub>O) is the principal gaseous N loss mechanism during nitrification on soil, and (4) the biogenic production of NO is so closely related to that of N<sub>2</sub>O that the soil emission of either gas cannot be understood at the process level without simultaneous measurement of the other, it follows that there is urgent need to characterize both the magnitude and direction of soil and foliar gaseous NO<sub>x</sub> exchange on agronomically - and environmentally - important situations, to determine the relationship of NO to N<sub>2</sub>O emissions on each situation, to identify factors involved on the control of these two processes, and to assess their importance to crop productivity, to N use efficiency, and to the long-term behavior of natural and other low-N ecosystems.
2. Because there is increasing public concern regarding (1) the agro-ecological and socio-economic impacts of threatened global climate change, and (2) additional potential health effects of changing atmospheric ozone concentration, and because gaseous N oxides are involved on "greenhouse warming" as well as ozone production/destruction, then it follows that there is urgent need to determine agriculture's contribution to global NO<sub>x</sub> and N<sub>2</sub>O budgets as well as to determine the "feedback" effect of both impending changes on the chemical and biological processes that collectively determine N availability, N use efficiency, and N contamination of the environment.

## Current Research Activities of G. L. Hutchinson

Numerous articles in both the popular and scientific press signal increasing public concern regarding not only the agro-ecological and socio-economic impacts of impending global climate change, but also additional potential health effects of changing atmospheric ozone concentration. Because gaseous N oxides are involved in "greenhouse warming" as well as ozone production/destruction, and because microbial processes in soil is one of the principal sources of atmospheric N oxides, then it becomes important to determine the magnitude of this source and, if appropriate, to develop control technology. During the last decade numerous measurements of soil N<sub>2</sub>O emissions have enhanced understanding of the factors controlling this process and of the importance of soil emissions compared to other sources of this gas. My research focuses instead on NO<sub>x</sub> exchange by soils and plants, for which very few measurements have been made. In addition to their important impacts on the chemistry of the atmosphere, we have wondered if soil NO<sub>x</sub> emissions might explain a significant fraction of the unaccounted-for N losses frequently observed in soil N balance sheets, and if NO<sub>x</sub> might serve, like NH<sub>3</sub>, as an agent for the transport and redistribution of N both within and among natural and agricultural ecosystems.

Overall long-term objectives of this research are to characterize both the magnitude and direction of soil and foliar gaseous NO<sub>x</sub> exchange in a variety of agronomically- and environmentally-important situations, to determine the relationship of NO<sub>x</sub> to N<sub>2</sub>O emissions in each situation, to identify factors involved in the control of these two processes, and to assess their importance to crop productivity, to N use efficiency, and to the long-term behavior of natural and other low-N ecosystems. Results of field measurements indicate that soil NO<sub>x</sub> emissions may be higher than commonly believed, that natural (rather than agricultural) ecosystems may have greater potential for aerobic loss of N oxides, and that emission of NO (rather than N<sub>2</sub>O) is the principal gaseous N loss mechanism during nitrification in aerobic soil. In the laboratory we used selective inhibitors and a glucose amendment to demonstrate that chemoautotrophic NH<sub>4</sub><sup>+</sup>-oxidizers (rather than autotrophic NO<sub>2</sub><sup>-</sup>oxidizers or heterotrophic nitrifiers) were primarily responsible for NO and N<sub>2</sub>O emissions from aerobically-incubated soil following addition of an ammoniacal N amendment. Other ongoing incubation studies involve using selective inhibitors and an O<sub>2</sub> concentration variable to characterize total NO emissions and the NO:N<sub>2</sub>O product ratio of various microbial N transformation processes. In cooperative research conducted at PrairieView A & M University, measurement of NO and N<sub>2</sub>O emissions from Bermuda grass plots follows a sampling schedule designed to determine the importances of precipitation, diel temperature fluctuation, seasonal change, and management (fertilization, harvest, etc.) as drivers of N oxide emissions. To help determine the relative importances of nitrification, denitrification, and chemo-denitrification as sources of the measured N oxides, we conduct detailed laboratory incubation studies of soil from selected field plots and perform analyses of periodic soil samples, as well as buried bags of soil, to determine each plot's NH<sub>4</sub><sup>+</sup>, NO<sub>2</sub><sup>-</sup>, and NO<sub>3</sub><sup>-</sup> pool sizes and transformation rates. Results indicate that NO and N<sub>2</sub>O arise primarily during biological (rather than chemical) transformations of soil N, that nitrification is probably a more important source of this gas than denitrification, and that the biogenic production and emission of NO is so closely related to that of N<sub>2</sub>O that the emission of either gas cannot be understood at the process level without simultaneous measurement of the other. We are developing a model that combines diurnal and episodic (in response to fertilization and/or precipitation) measurements with periodic background measurements to estimate the annual NO and N<sub>2</sub>O flux from subtropical grassland ecosystems, which apparently make a disproportionately large contribution to global NO and N<sub>2</sub>O budgets.

## FUTURE PROBLEMS AND RESEARCH PRIORITIES FOR NITROGEN RESEARCH IN ARS

A. R. Mosier, Soil-Plant Nutrient Research  
Fort Collins, Colorado

Agricultural production and environmental issues are inseparable when N is concerned. Being an essential and generally limiting element for crop production, it is necessary to add N to most crops. The amount of N that crops most efficiently use, the economics of adding excess N, and environmental aspects of N leakage from agricultural systems is not in balance. We are therefore faced with the problem of producing an adequate quantity of food and fiber at economically viable expense without harming our local and global environment. Inefficient use of fertilizer N can result in contamination of ground water supplies through nitrate percolation, excessive denitrification and  $N_2O$  production, alteration of  $N_2O/N_2$  ratios, and interactions which create imbalance in other nutrient cycles. Since agriculture is a large consumer of energy and water resources, methods of managing agriculture must be continually critically assessed. Nitrogen research priorities to address the role of N in the interactions of production agriculture and environmental quality should include the following:

1. Improving N fertilizer use efficiency.
2. Determine the role of agriculture in biogenic trace gas ( $N_2O$ ,  $NO_x$ ,  $CH_4$ , hydrocarbons, sulfur compounds) production. Gain knowledge about trace gas emission rates from existing cropping systems.
3. Determine the effect of changing agricultural technology on trace gas production.
4. Effect of global climate change on U.S. agriculture.

## CURRENT RESEARCH ACTIVITIES

A. R. Mosier, Soil-Plant Nutrient Research  
Fort Collins, Colorado

1. Field quantification of denitrification in agricultural systems.
2. Measurement and modeling of nitrous oxide emissions from native and agricultural systems.
3. Plant rhizosphere-N transformations interactions.
4. Development of encapsulated calcium carbide as a nitrification inhibitor.
5. Effect of N-transformation inhibitors on:
  - a. Fertilizer N use efficiency.
  - b. Nitrous oxide production from fertilizer N.
  - c. Nitrate leaching.
6. Production and consumption of methane in native and agricultural systems and predictive model development.

**USDA-ARS Working Conference on  
Nitrogen Research**

**D. R. Timmons, Soil Scientist  
National Soil Tilth Laboratory  
Ames, IA**

**Current Research Activities**

- I. Use of point-injection fertilizer technology to improve fertilizer N management.
  1. Determine whether ammonium-N, nitrate-N, or a ratio of these mineral N forms is preferred by corn when injected at different growth stages.
  2. Evaluate injecting a nitrification inhibitor with N solutions containing ammonium-N at different corn growth stages to maintain the N in a nonleachable form for a longer duration.
  3. Develop fertilizer nitrogen management that will increase crop uptake and minimize nitrate-nitrogen losses below the root zone by using point-injection technology for more precise placement and timing of applied nitrogen.
  
- II. Identify and quantify soil N x crop x tillage interactions so residual soil N transformations and fertilizer N application can be synchronized with plant N requirements.
  1. Determine the fate of injected tagged fertilizer N in soil environments and crop residue distributions created by different tillage practices.
  2. Develop fertilizer N and crop management practices that synchronize residual soil nitrate and seasonal soil N mineralization with fertilizer N application to satisfy corn N requirements.

USDA-ARS Working Conference on  
Nitrogen Research

D. R. Timmons, Soil Scientist  
National Soil Tilth Laboratory  
Ames, IA

Future N Research Needs

1. Identify and quantify soil N interactions among chemical and biological processes resulting from tillage and cropping systems that affect N availability, N transformations, and N leaching losses.
2. Develop rapid and accurate methods for measuring and predicting potentially mineralizable soil N throughout the growing season so fertilizer N applications can be synchronized with this N source and with residual soil nitrate-N to meet crop N requirements.
3. Develop rapid and accurate methods for sampling and measuring residual soil nitrate-N status for N management systems other than surface-applied N. This would include point-injected N and banded (knifed-in) N techniques used to apply N solutions, anhydrous ammonia, animal wastes, sludge, etc.
4. Evaluate using cover crops (particularly legumes) as a N source for potentially mineralizable soil N and as a N sink for utilizing nitrates below the normal rooting depths of major crops.
5. Develop improved techniques for determining denitrification under field conditions for soil environments and residue distributions created by different tillage systems.
6. Determine the interactions among biological, chemical, and physical processes involved in forming, maintaining, and destroying soil aggregates to understand how soil structure affects N movement in the soil solution.

**WORKING GROUP 6**  
**NITROGEN NUTRITION OF PLANTS**





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ARS Working Conference on Nitrogen Research.

## 1. Research Related to Nitrogen Nutrition of Plants:

I have been conducting research on free-living (or associative), nitrogen-fixing bacteria in the rhizosphere of plants in a terrestrial environment. The long-term agronomic objective is to provide increased nitrogen for crop growth and yield at a reasonable cost. The research goals are to quantify the amount of nitrogen fixed, determine the amount of the fixed nitrogen that is available for the plant use and then identify methods to increase this nitrogen source.

To augment the rather low levels of nitrogen fixed by "natural" associations, inoculation investigations were initiated. Inoculation experiments in the field and greenhouse have been extremely variable. Some combinations of plants and nitrogen-fixing bacteria show a positive response (increased yield, either grain or forage) to inoculation, others exhibited no measurable response and in some combinations inoculation was detrimental. In addition, the results can be variable in time and/or location (eg. a positive response one year with a particular combination could be negative the next year at the same location using similar management practices). The variability of the association and the low levels of plant response and/or nitrogen fixation after inoculation promoted an investigation into the possible causes.

Substantial numbers of metabolically active bacteria are required to provide significant quantities of nitrogen. However, we find that, in most instances, there is the rapid loss of large numbers of the inoculated microorganisms from the rhizosphere. As many as  $10^5$  nitrogen-fixing bacteria can be lost during the first week following inoculation. This loss, although reduced, generally continues during the ontogeny of the plant. This loss is exacerbated by several stress factors, including suboptimal soil pH, temperature and moisture. The inoculated populations are not lost as rapidly in a "sterile" soil when compared to a "non-sterile" soil indicating that competition and/or predation is a major factor in the removal of the nitrogen-fixing bacteria. We have identified several bacterial predators from inoculated soils. Several attempts to overcome this rapid decrease in inoculated nitrogen-fixing bacteria have not been successful.

In addition to large bacterial populations, effective associations require an ample supply of readily metabolizable reduced carbon substrates. This is needed, not only for the growth of the bacteria, but to supply energy and reductant for nitrogenase activity. Our interpretation of results using either total nitrogen accumulation or the acetylene reduction technique is that these associations do not produce significant (above 10 to 20 kg N/acre when extrapolated to an

acre basis) quantities of nitrogen. Our investigations show that the amount of nitrogen that can be fixed by these systems, based on root exudation of recent photosynthates, is generally in the range of 2 to 12 Kg N/acre. This is in general agreement with many nitrogen balance studies, however, it is far less than many projections made using the acetylene reduction technique. We have been able to stimulate nitrogen fixation, as determined by  $^{15}\text{N}_2$  incorporation into the soil or plant material by adding sugars and organic acids, either alone or in combinations, to the rhizosphere. This strongly suggests that there is an inadequate energy supply in the rhizosphere to make this system reliably produce significant quantities of nitrogen.

## 2. Primary Issues Facing ARS in this Area:

Should we hold out any hope that associative nitrogen fixation can contribute a significant amount of nitrogen to an agricultural system in the US? My response is a qualified yes. In some forests, grasslands and aquatic environments, including rice production, associative nitrogen fixation may make a substantial contribution to the nitrogen economy of the ecosystem. Should this area receive support? Despite the rather negative tenor of the previous section, I believe research in this area should be continued. At the basic level, the study of these organisms has been most rewarding. Most of what we know about the biochemistry and genetics of nitrogenase and nitrogenase reductase came from non-symbiotic, or associative, organisms (eg. Klebsiella pneumoniae & Azotobacter spp.). Large scale inoculation or "cut and weigh" type studies are not productive at this time, however, basic investigations in this area should be strengthened.

## 3. Future Research Imperatives.

Future research should utilize productive associations (eg. aquatic) to understand the basic biology of the systems, rather than attempting to force unproductive ones to function beyond their present capacity. A better understanding of the biology of bacterial partner is required. More basic research in this area is badly needed. Investigations on the molecular biology and enzymology of nitrogen fixation and assimilation in these organisms should be continued and support for these studies increased. It is this type of research that will eventually lead to transferring nitrogen-fixing capability to higher plants. The ecology of nitrogen-fixing bacteria in the rhizosphere should be supported as well. The information gathered from this type of work could be useful in other areas, eg. biological control, plant pathology, and the stimulation of crop growth by beneficial rhizosphere bacteria, regardless of the mechanism.

I would like to suggest that transferring the genes for nitrogen fixation from a prokaryotic to eukaryotic organism and obtaining a functional gene product should be a research imperative for ARS. This seems like a good project for (a) federal lab(s). It is the logical extension of much of the nitrogenase enzymology and molecular biology research, such crops would be of great value for American agriculture and the project is probably beyond the scope of most labs in private industry.

Breeding for Improved N<sub>2</sub> Fixation and N Nutrition in Alfalfa<sup>1</sup>  
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Accomplishments

Breeding for increased biological N<sub>2</sub> fixation requires the simultaneous improvement of many plant characters. Research has been conducted on this topic for 15 years at St. Paul, MN. Our multidisciplinary research team has used bidirectional selection to produce alfalfa subpopulations with high and low levels of nodulation, fibrous root development, herbage weight, root weight, root N concentration, three nodule enzymes [nitrogenase (NA), phosphoenolpyruvate carboxylase (PEPC), and glutamate synthase (GOGAT)] and leaf nitrate reductase (NRA). Comparative studies of the various high, low, and unselected subpopulations have provided information about the effect of each trait on N<sub>2</sub> fixation, associated traits, and yield.

Selecting for increased nodule mass and increased plant size traits generally increased N<sub>2</sub> fixation by increasing nodule mass. However, more than half of the variation in N<sub>2</sub> fixation was considered to be host genotype x Rhizobium strain effects. The most effective symbiotic combinations are those where highly nodulated genotypes are infected by highly effective Rhizobium strains. A multiple-step breeding scheme was conceived to enhance N<sub>2</sub> fixation by selecting for the improvement of coordinated responses. Large populations of plants are grown in nil-nitrate soil inoculated with both highly effective and low effectiveness Rhizobium strains. At four weeks of age the 75% least vigorous seedlings are discarded. At twelve weeks plants are observed in groups of about 25. The 10-12 best plants, (based on large herbage mass, large root mass, and large nodule mass) are selected and evaluated for NA. The best 5 or 6 plants per group are saved based on NA. The goal is to save 120-200 plants per germplasm per cycle of recurrent selection. This selection scheme has been used successfully to improve populations for plant characters associated with N<sub>2</sub> fixation and to improve host preference for highly effective rhizobial strains. This selection scheme also increases activities of the nodule nitrogen and carbon assimilating enzymes, GOGAT and PEPC.

Populations obtained from the multiple-step selection program often fixed more N, but did not always produce increases in forage and N yields, especially in the presence of large amounts of soil N. We have hypothesized that some of the problems were caused by: inbreeding effects, a need for increased N transport out of the nodule to the herbage and roots, and a need for plant and Rhizobial genotypes that can fix N in the presence of high soil nitrate-N.

The inbreeding effects caused by selecting for coordinated responses of several plant characters have been reduced by using several heterogenous germplasm sources as base populations. Selection is conducted independently in each germplasm and then two germplasms are strain crossed, thus producing heterosis for forage yield in addition to improved N<sub>2</sub> fixation.

We have conducted a bidirectional selection study to develop four basic types of populations: high nodule enzyme (GOGAT and PEPC) + high N transport enzyme [aspartate amino transferase (AAT) and asparagine synthetase (AS)], high nodule enzyme + low transport enzyme, low nodule enzyme + high transport enzyme, and low nodule enzyme + low transport enzyme. These populations have been developed, but the effect of selection on plant performance remains to be determined.

N<sub>2</sub> fixation in the presence of high soil nitrate-N has shown very little plant response. However, R. meliloti isolates have been identified that are tolerant to soil nitrate-N concentrations that normally affect the symbioses of most indigenous and inoculum strains.

We concluded that selection for increased N<sub>2</sub> fixation should be accompanied by selection for yield and quality traits that utilize the increased capability to symbiotically fix N. We conducted two cycles of selection in two nondormant germplasms for increased amounts of forage, crowns, and roots during autumn, and for increased concentrations of N stored in the tap root. The strain cross between the two selected germplasms produced the new cultivar, 'Nitro'. Nitro provides about 60% more N (43 vs. 27 k) in the seeding year for plow-down than dormant check cultivars. In addition to showing greater root yields and increased concentrations of root-N, there was a 7 percent improvement in nitrogen fixing ability for Nitro over the parental germplasms. The N<sub>2</sub> fixation research group at the University of California-Davis, lead by L.R. Teuber and D.A. Phillips, have successfully selected for increased dry matter production and reduced-N concentration in alfalfa forage.

Non-N<sub>2</sub> fixing (non-nodulation and ineffective nodulation) germplasms were developed for use in basic nodule development research and for use as controls in estimating N<sub>2</sub> fixation in field experiments with both the <sup>15</sup>N isotope dilution method and by the difference method. These populations have been increased and made available to scientists for use in routine trials.

#### Primary Issues for ARS Researchers

Nodule mass is a critical factor affecting N<sub>2</sub> fixation. We need a better understanding about how soil organisms (i.e., bacteria, fungi, nematodes, and insects) affect nodulation and nodule function. There is a need to study the affect of crop management on N<sub>2</sub> fixation and its effect on crop quality. Some Rhizobium research needs to be maintained. This includes development of an international 'core' collection and microbial genetic research to improve the N<sub>2</sub> fixation capacity of strains adapted to specific regions, especially those with problem soils, such as high Aluminum and low pH. The available R. meliloti collection housed at St. Paul needs to be stored, maintained, and distributed from a central location, possibly Beltsville.

#### Future Research Imperatives

A simplified alfalfa breeding protocol needs to be developed that combines all of the existing selection methodology and knowledge of alfalfa N<sub>2</sub> fixation. This endeavor is necessary if there is to be N<sub>2</sub> fixation technology transfer to the alfalfa breeding industry. This may require an announced project to breed high N<sub>2</sub> fixing alfalfa cultivars with increased yield and forage quality and adapted to the midwest United States.

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<sup>1</sup> Prepared for the "ARS Working Conference on Nitrogen Research," St. Louis, MO, May 22-25, 1989.

Summary of Research Interests  
(P. E. Bishop)

In 1980 we reported evidence for the presence of an alternative  $N_2$  fixation system in the soil bacterium Azotobacter vinelandii which is expressed in the absence of molybdenum. Since molybdenum has long been thought to be essential for nitrogen fixation, this was a rather startling finding which caused much initial skepticism. Since that time nifHDK (structural genes encoding the conventional molybdenum-containing nitrogenase) deletion strains have been used to provide definitive evidence for the existence of two alternative nitrogen fixation systems in A. vinelandii. One of the alternative nitrogenases is a vanadium-containing enzyme complex while the other appears to be devoid of both vanadium and molybdenum. We have concentrated our efforts on the latter enzyme system since this nitrogenase may ultimately provide the best means to a more complete understanding of how nitrogenases catalyze the reduction of  $N_2$ . Our ultimate objectives are to understand the mechanism of the  $N_2$  reduction by this new nitrogenase (the second alternative nitrogenase) and the genetic organization of both this system and the vanadium enzyme system at the nucleotide level. Our approaches involve the use of anaerobic columns to purify the protein components of this new nitrogenase and both chemical and physical procedures to characterize these proteins. We have cloned DNAs containing a number of the genes involved in this system and have sequenced many of these genes using the dideoxy chain-termination method. We have inserted kanamycin-resistance ( $Kan^r$ ) cartridges into unique restriction enzyme sites located in open-reading frames (ORFs) deduced from the nucleotide sequence data. The mutants generated by inserting the  $Kan^r$  cartridges into these ORFs are being characterized with respect to their nitrogen-fixing (Nif) phenotype in the presence of molybdenum or vanadium and the absence of either metal. Northern blot experiments are being conducted on these mutants to determine the absence or presence of mRNAs transcribed from the mutated genes. We also plan to further investigate the regulation of alternative nitrogenases by molybdenum and vanadium using a combination of biochemical and molecular biology techniques. Finally, we would like to begin to ask questions about the distribution of alternative  $N_2$  fixation systems in other bacteria (besides A. vinelandii) and the importance of these systems to the global nitrogen cycle. Answers to these questions might also lead to insights on why bacteria have alternative  $N_2$  fixation systems. On the more practical side, transfer of genes encoding alternative  $N_2$  fixation systems to agronomically useful bacteria such as rhizobia (assuming the absence of alternative  $N_2$  fixation systems in rhizobia) might provide some benefit to leguminous crops cultivated on molybdenum-deficient soils.

Report on Individual Research Accomplishments,  
Primary Issues Facing ARS in the Area of Plant N Nutrition, and  
Future Research Imperatives

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I. Individual Research Accomplishments:

- A. The Identification of Genes in Soybean that Restrict Nodulation of Highly Competitive Indigenous but Relatively Ineffective *Bradyrhizobium japonicum*. In soybean grown in the midwestern US, most nodules are occupied by *B. japonicum* strains belonging to serocluster 123. Based upon the analogy to a host-pathogen interaction, i.e., that genes for "resistance" to nodulation by certain strains in the soybean host function in a "gene-for-gene" fashion with complementary genes in *B. japonicum* (U. S. Patent Application Serial No. 07/087,356) we identified soybean lines that resisted nodulation by strain USDA 123 (1). These genotypes, typified by PI 377578, reduced the competitiveness of USDA 123 and certain other serocluster 123 isolates in favor of inoculant quality strains (3, 11). We determined that a relationship existed between genomic DNA *Eco*R1 digestion pattern, SDS-protein profile, and Southern hybridization to a *nif*H probe and the ability of a strain to nodulate PI 377578 (13). Inheritance studies of restricted nodulation of USDA 123 indicate that this trait is controlled by a single dominant allele in PI 377578 (7). We have recently isolated a corresponding *B. japonicum* gene that allows nodulation of PI 377578 (14).

Because PI 377578 restricted only a portion of the serocluster 123 strains tested, we undertook further studies to identify soybean germplasm that resisted nodulation by serocluster 123 strain MN1-1c. PI 417566 and PI 283326 were found that resist nodulation by and reduce the competitiveness of strain MN1-1c (6). The gene in PI 417566 that restricts nodulation of strain MN1-1c is inherited as a single dominant (5). Of 20 serocluster 123 strains collected from around the US and the PRC, the nodulation of only three is not restricted by at least one of the three soybean genotypes, PI 377578, PI 417566, or PI 283326 (4).

We have also attempted to exploit a genetic system whereby the soybean host genotype eliminates nodulation by indigenous serocluster 123 strains in favor of the fast-growing soybean microsymbiont, *Rhizobium fredii*. We identified a *Glycine soja* (wild soybean) genotype that nodulates primarily with *R. fredii* USDA 193 when in competition with strain USDA 123 (2). This is in contrast with the standard soybean cultivar Williams, that forms 100% of its nodules with USDA 123 under the same conditions.

I would expect that I will have agronomically adapted materials available for field testing in about five years with the possibility of germplasm release shortly thereafter. Because a patent has been applied for to protect this approach to reducing nodulation by indigenous rhizobia, I have also had conversations with a number of commercial firms interested in cooperative efforts.

- B. Heterosis of Interspecific Soybean Hybrids for Traits Related to N<sub>2</sub> Fixation  
In an effort to find novel sources of soybean germplasm and thus allow greater genetic variation for N<sub>2</sub> fixation, we identified desirable wild soybean genotypes and made interspecific hybrids with selected soybean cultivars. F<sub>1</sub> hybrids were tested which, on the average, exceeded the cultivated soybean parent by 34 and 28%, respectively, for nodule mass and N accumulation (9). We are now attempting to select segregants with enhanced N<sub>2</sub> fixation from certain of these crosses.

- C. N Accumulation and Partitioning in Soybean Germplasm - Based upon a scheme proposed earlier (8) we studied the N accumulation and partitioning of soybean cultivars and germplasm accessions (10). Modern cultivars generally showed maximal N accumulation and partitioning suggesting that these traits were associated with agronomic improvement. No germplasm accessions were identified that significantly exceeded the modern cultivars in N accumulation or partitioning. More recently, we have attempted to determine differences in the N metabolism of a normal and a high percent seed protein soybean genotype which contributed to the high seed protein of the latter (12). Higher seed protein was associated with greater  $N_2$  fixation, and superior N partitioning.

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#### II. Issues Facing ARS in the Area of Plant N Nutrition

- A. An issue of overriding importance in all areas of ARS funded research is the provision of adequate funding levels to provide ARS research projects levels of manpower and support to attract and keep highly qualified scientists and to compete with well funded state programs.
- B. The integration of  $N_2$  fixation research with research related to other aspects of plant N metabolism.

#### III. Future Research Imperatives

- A. Development of superior legume host/rhizobial combinations through the simultaneous genetic alteration of both partners of the symbiosis.
- B. Multifaceted approach toward the displacement of competitive but ineffective indigenous rhizobia with highly effective inoculant strains. Approaches might include 1) superior host-rhizobium specificity, 2) co-inoculation with other microorganisms, 3) use of soil applied chemicals, and 4) enhanced inoculant delivery techniques.
- C. The identification and characterization of plant genetic factors affecting  $N_2$  fixation, N metabolism, and rhizobial specificity.
- D. The development of plant genotypes that produce unique seed or other plant tissue protein products which can be produced for speciality markets.

## CONTRIBUTION TO "NITROGEN NUTRITION OF PLANTS" WORKING GROUP

J.E. HARPER, URBANA, IL.

### BACKGROUND AND ACHIEVEMENTS

The ability of grain legumes to symbiotically fix atmospheric  $N_2$  has provided a mechanism for these species to survive on N-deficient soils. Much of the soybean production area in the U.S., however, involves soils which have appreciable levels of residual N and, thus, the symbiotic  $N_2$ -fixation system is functioning at less than maximum rates, due to inhibitory effects of nitrate. In spite of the fact that inhibition of nodulation by nitrate has been known for many years, the mechanism(s) of this inhibition remains elusive and thus efforts to circumvent this inhibition have not been successful to date. Work at Urbana did demonstrate that complete dependence of the soybean plant on dinitrogen did not support optimum growth and seed production (Harper, 1974). Thus, there appears to be limits on the extent to which one can feasibly enhance the plants dependence on  $N_2$  fixation.

With regard to mechanism of nitrate inhibition of nodulation, one hypothesis is that nitrate metabolism effectively competes for a limited photosynthate pool, which in turn limits carbon availability for support of nodule growth and function. It has been concluded that the soybean host plant rather than the bacterial microsymbiont is in primary control of nodulation response to nitrate nutrition (Gibson and Harper, 1985). Under this premise, effort was directed to selection of soybean lines with limited nitrate metabolism capabilities; more specifically we sought lines which lacked the nitrate reductase (NR) enzyme. The approach taken was to induce genetic variability for this trait through mutagenesis (Ryan and Harper, 1983). Mutant lines were isolated with altered NR activity, and it was ascertained that soybean contains multiple NR isoforms. The initial mutant isolated lacked both constitutive NR isoforms; the NAD(P)H, pH 6.5 enzyme and the NADH, pH 6.5 enzyme (Nelson et al., 1983). This mutant is controlled by a single recessive gene, designated  $nr_1$ , and has been lodged in the soybean genetic type collection as T276. The  $nr_1$  line retains the NADH (pH 7.5) substrate-inducible NR form and is not altered in its ability to take up and metabolize nitrate, nor is it altered in terms of nitrate inhibition of the nodulation process (Ryan et al., 1983). It is known that the substrate-inducible NR isoform is the dominant NR isozyme in soybean and it appears that the level of this enzyme is adequate to metabolize all incoming nitrate. Hence, the overall metabolism of nitrate and dinitrogen is unaffected in the  $nr_1$  mutant.

Two additional mutants have been isolated which lack one of the two constitutive NR isoforms (Streit and Harper, 1986). These mutants are also unchanged with respect to nodulation response to nitrate, but have served a very useful purpose in allowing verification that the pH 6.5 NAD(P)H enzyme is responsible for a secondary metabolic pathway in soybean which converts nitrite to a gaseous form, primarily nitric oxide (Dean and Harper, 1988). This pathway is not, however, of any major concern to the overall nitrogen use efficiency of soybean since there is an a priori requirement that nitrite accumulate in the tissue before there is any resulting gaseous losses; a condition that rarely, if ever, occurs in intact plants in their normal growth habit.

We have been unsuccessful in selecting a line which lacks the NADH, pH 7.5, NR isoform and as such have not been able to demonstrate whether blocking NR activity would allow nodulation to occur in the presence of nitrate. The NR mutants which have been isolated have provided excellent sources of materials from which to purify and biochemically characterize the NR isoforms and have greatly extended our scientific knowledge concerning genetic control and biochemical properties of the constitutive and inducible NR isozymes (Harper, 1988).

We have also successfully selected soybean lines from mutagenized populations which have enhanced nodulation (hypernodulating lines) in the absence and presence of nitrate (Gremaud and Harper, 1989). These mutants are altered in the autoregulatory control of nodule number and have three to five times greater numbers of nodules than does the Williams parent line. The hypernodulating lines are controlled by a single recessive gene in three of the mutants tested. The infection and initial nodulation development stages are partially tolerant to nitrate, while functional nodule activity (nitrogenase) does not appear to be tolerant of nitrate. An additional mutant line has been selected which does not nodulate when challenged with Bradyrhizobium japonicum. This line will be beneficial in tests to evaluate  $N_2$  fixation by the difference method. Based on grafting studies, the shoot exercises



control over the greater nodule numbers of the selected hypernodulating mutants, while the root exercises control over the nonnodulating trait (Harper, unpublished). Initial yield tests indicate that Williams yields more than the hypernodulating mutants (Wu and Harper, unpublished). This was not unexpected inasmuch as the mutational approach likely resulted in multiple mutations in addition to the nodulation trait on which selection was based. A backcross program has been initiated in an attempt to move the hypernodulation trait into a background closer to the Williams parent and these lines should be available for public release in 3-5 years if they have desirable agronomic responses.

### PRIMARY ISSUES

1. Continued effort to enhance symbiotic N<sub>2</sub> fixation by soybean, with the view that companion crops or subsequent crops grown in rotation may benefit from greater residual plant-available soil N, remains as a viable research area.
2. The major deterrent to enhanced nodulation in the Mid West is the inhibitory effect of soil nitrate on nodule initiation and functional activity.
3. Nitrate uptake, rather than level of NR activity, appears to be the primary limitation to nitrate metabolism in soybean, and likely in most crops.

### RESEARCH IMPERATIVES

1. Effort should continue to ascertain the mechanism of nitrate inhibition of nodulation and seek ways to enhance dinitrogen fixation in the presence of nitrate. Mutagenesis and molecular biological techniques provide new approaches to this problem area. The goal is to develop lines with enhanced N<sub>2</sub> fixation and to release lines to the public in the next 5 years.
2. Regulation of nitrate uptake carrier proteins should be characterized to allow manipulation of nitrate uptake and transport within the plant. Progress will be dependant upon successful identification of carrier proteins.

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## RECENT CONTRIBUTIONS

Gary H. Heichel  
St. Paul, MN

1. Obtained first direct measurements of inter- and intraannual patterns of  $N_2$  fixation in field communities of perennial forage legumes. This information revealed how  $N_2$  fixation changed with species, with plant population, with shoot and root growth within and across years, and with harvest management.
2. Conceived and implemented first field scale methods (N-15 isotope dilution and difference techniques) for evaluation of perennial legume germplasm in situ for improved  $N_2$  fixation. Proved field performance of the first non- $N_2$  fixing alfalfa germplasm selected and released, and of the first crop variety (an "annual" alfalfa) developed and released for enhanced N accumulation and storage characteristics.
3. First use of N-15 methodologies to demonstrate in the field that the legume host plant capability for  $N_2$  fixation could be repeatedly distinguished from rhizobial and environmental influences. This contributed to the development of a new and more efficient program for simultaneous host-rhizobial selection that has application to  $N_2$  fixation breeding efforts on many crop legumes.
4. Conceived and developed the first quantitative N budgets of forage legume crops by use of  $^{15}N$  methods. These budgets established how to manage legumes for net return of fixed  $N_2$  to the soil-crop system, and permitted identification of shortcomings in the standard fertilizer N equivalency concept of crediting legume N benefits to a subsequent crop.
5. Conducted first definitive field investigations in North America of the occurrence and significance to the donor (legume) and recipient (grass) of transfer of symbiotically-fixed N from forage legumes to a forage grass. N transfer occurred over distances as great as 20 cm, and accounted for up to 79% of the N accumulated by the grass.

## CURRENT RESEARCH OBJECTIVES

Gary H. Heichel  
St. Paul, MN

1. Determine the importance of legume nodule  $\text{CO}_2$  fixation and other host/rhizobial traits to plant C economy and to efficiency of nodule  $\text{N}_2$  fixation.
  - a. Determine effect of host genotype and stage of development on C costs of  $\text{N}_2$  fixation.
  - b. Determine effect of rhizobial species and strain effectiveness on C costs of  $\text{N}_2$  fixation.
  - c. Isolate and purify nodule glutamate synthase, and produce polyclonal antibodies to enzyme for further fundamental studies.
2. Determine the importance of N transfer from legumes to nonlegumes on crop community productivity and persistence (N-15 methods).
  - a. Measure legume-grass N transfer in stands with defined interplant geometries.
  - b. Measure legume-grass N transfer over 4 year stands of major forage legumes with grass.
  - c. Determine recovery of legume N by succeeding nonlegume crop and loss of legume N in leachate from rooting zone.
3. Conceive and implement methods for field scale measurements of  $\text{N}_2$  fixation for use in legume germplasm improvement programs and plant-soil N cycle research (N-15 methods).
  - a. Determine  $\text{N}_2$  fixation of experimental alfalfa germplasm selected for differences in nitrate reductase activity.
  - b. Determine suitability of ineffectively nodulated alfalfas as controls in isotope dilution and difference method approaches to measuring  $\text{N}_2$  fixation.
  - c. Test suitability of N-15 isotope dilution and difference methods for measuring  $\text{N}_2$  fixation of minor use forage legumes.

## FUTURE RESEARCH PRIORITIES

Gary H. Heichel  
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### A. PHYSIOLOGY AND BIOCHEMISTRY OF N<sub>2</sub> FIXATION.

1. Determine whether C costs of symbiotic N<sub>2</sub> fixation limit dry matter accumulation of nodulated legumes.
2. Determine whether availability of C to the bacteroid limits symbiotic N<sub>2</sub> fixation in situ.
3. Determine response of legume N assimilation and partitioning to incremental CO<sub>2</sub> enrichment and anticipated associated stresses.
4. Determine mechanisms of regulation of enzyme activity for key root nodule enzymes with activities modified by selection by traditional (Mendelian) genetic methods.

### B. NITROGEN CYCLING IN CROPPING SYSTEMS

1. Determine extent and significance of losses of legume N to groundwater.
2. Determine factors that regulate the effectiveness of alfalfa and other forage legumes in recovering excess subsoil nitrate-N.
3. Determine the fate of fertilizer and legume N at a network of geographical sites that may pose risks to groundwater. Obtain complete N balances at all sites using identical methods for fertilizer N-based and legume N-based cropping systems.

Enhanced symbiotic nitrogen fixation by Bradyrhizobium japonicum strains with altered tryptophan metabolism

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The overall objective of my research project has been to develop methods to enhance the agricultural use of N-fixation. Increased agricultural use of N-fixation is desirable as it reduces the farmers need for chemical nitrogen fertilizers, thus reducing operating costs while preserving energy resources and protecting our water resources from fertilizer run-off. Recent research investigations have examined the role bacterial tryptophan biosynthesis plays in the Bradyrhizobium japonicum - soybean symbiotic association. There are several reasons to suspect that a product or products from this pathway may influence the symbiotic properties of the microsymbiot. First, bradyrhizobia with altered tryptophan catabolism appear to have altered symbiotic properties (1). Also, mutant bradyrhizobia (auxotrophs) with defects in their tryptophan biosynthetic pathway often cannot nodulate (2). These observations suggest that a bacterial product made from tryptophan or made from a tryptophan pathway intermediate is involved in the nodulation process. The indole compounds that act as plant hormones are, of course, prime candidates. Bacteria, including rhizobia and bradyrhizobia, can really convert tryptophan to indole compounds such as indoleacetic (IAA), indolelactic (ILA) and indolepyruvic acid (IPyA). Many of these indole compounds can impact the development and functioning of plant structures, and it seems reasonable to suspect that the production of these compounds by rhizobia or bradyrhizobia could influence the symbiotic process. Thus, it may be possible to improve the legume-Bradyrhizobium (-Rhizobium) symbiotic association by modifying the microsymbiot so that optimal amounts of these hormones are produced. Recent work has looked at the effect such altered bacteria have on symbiotic N-fixation.

The bacterial mutants that have been investigated include several B. japonicum mutants with altered tryptophan metabolism resulting from spontaneous resistance to 5-methyltryptophan (the 5-MT mutants) and a prototrophic revertant of a B. japonicum tryptophan auxotroph (the NOD+ mutant). The NOD+ mutant was isolated by Dave Kuykendall, USDA-ARS, Beltsville and Dave and I are cooperating in studies with this mutant. The wild-type for both the 5-MT and NOD+ mutants is B. japonicum I-110 ARS.

Greenhouse studies, conducted with the 5-MT mutants, demonstrate that the symbiotic properties of the legume-Bradyrhizobium association can be changed by using inocula with altered tryptophan biosynthesis. These studies show a variety of effects depending upon the mutant strain used (Table 1). With the 5-MT-3 strain nodule number remained at the wild-type level but nodule mass decreased and when the 5-MT-7 strain was used nodule number and mass were both dramatically decreased. With both the 5-MT-3 and -7 strains plant nitrogen content and dry weight decreased. In contrast, plants inoculated with the 5-MT-4 strain had more nodules and more nodule mass than did control plants inoculated with the wild-type (I-110 ARS) bacteria. Slight increases in plant dry weight and nitrogen content were even observed with this mutant. Nodule specific activity (N-fixed per gram of nodule) decreased in all plants receiving the 5-MT mutants. Laboratory studies showed that tryptophan, tryptophan intermediate and tryptophan degradation compounds (IAA, ILA, and IPyA) accumulate in cultures of these 5-MT bacteria, evidence that the metabolism of tryptophan had been altered in these bacteria. The accumulations observed with the 5-MT-4

strain differed from those observed with the 5-MT-3 and -7 strains, indicating that the tryptophan metabolism of this strain had been altered in a different manner. Unfortunately, the 5-MT-4 mutant was found to be unstable and more detailed studies were not conducted.

Table 1. Effect of 5-MT mutants on plant dry weight, nodulation and nitrogen content.

Inoculum	Plant		Nodule	
	Dry wt. (g)	Nitrogen (g)	Number	Mass <sup>1</sup> (g)
Wild-type	4.7	0.138	151	2.2
5-MT-4	5.2	0.145	349	3.9
5-MT-7	3.0	0.064	21	1.1
None	1.9	0.023	0	0

<sup>1</sup>Wet weight of nodules per plant.

The NOD+ strain appears to be stable and several greenhouse studies have been conducted with this mutant. Preliminary work (data not shown) indicated that plants receiving this mutant had better color, higher dry weights and larger nodule mass than did control plants inoculated with wild-type bacteria. More detailed studies show that plants inoculated with the NOD+ mutant were 33% larger, contained 52% more nitrogen and 34% more carbon than did control plants inoculated with the wild-type strain (Table 2). Also, plants treated with the NOD+ mutant had 56% more nodules and 41% more nodule mass than did the control plants. Average nodule size and amount of N-fixed per gram of nodule were about the same with both inocula. These data suggest that the improvement in N-fixation observed with the NOD+ mutant was due to an increase in nodule mass that resulted from an increase in nodule number.

Table 2. Effect of the NOD+ mutant on plant dry weight, carbon content, nitrogen content and nodulation.

Inoculum	Plant			Nodule	
	Dry wt.	Carbon (g)	Nitrogen	Number	Mass <sup>1</sup> (g)
None	2.1	0.68	0.027	0	0
Wild-type	6.4	2.55	0.192	134	2.9
NOD+	8.5	3.43	0.277	209	4.1

<sup>1</sup>Wet weight of nodules per plant.

The physiological basis for this increase in nodulation is under investigation. Again, it is suspected that altered tryptophan biosynthesis is involved. There are several reasons for this suspicion. First, the auxotroph from which the NOD+ mutant was derived was a well defined tryptophan auxotroph

known to be defective in a single tryptophan pathway enzyme (2). The NOD+ is a spontaneous revertant of this auxotroph and it is therefore extremely unlikely that more than a single mutation was involved in the reversion. The change that did occur involves the tryptophan pathway as it corrected for the enzyme deficiency that existed in the auxotroph. Also, the NOD+ and wild-type strains differ in their sensitivity to toxic amounts of tryptophan and in their uptake of anthranilic acid (a key component of tryptophan). The NOD+ may also produce more IPyA than does the wild-type strain.

These studies are consistent with the hypothesis that a bacterial produced tryptophan pathway product is involved in the nodulation process. Also, this work has demonstrated that legume inocula with improved symbiotic properties can be developed by altering the tryptophan biosynthesis of the inoculum strain.

#### Literature cited

1. Kaneshiro, T. and W.F. Kwolek. 1985. Stimulated nodulation of soybean by Rhizobium japonicum mutant (B-14075) that catabolizes the conversion of tryptophan to indol-3yl-acetic acid. Plant Sci. 42:141-146.
2. Wells, S.E. and L.D. Kuykendall. 1983. Tryptophan auxotrophs of Rhizobium japonicum. J. Bact. 156:1356-1358.

## FUTURE PROBLEMS AND PRIORITIES

Two major areas of research that I feel should be priority research areas are 1) developing inoculum strains that can compete better with inefficient native rhizobia and 2) improving the ability of the legume-Bradyrhizobium (-Rhizobium) association to fix nitrogen. There are certainly broad areas of research and there are many ways that these problems can be attacked. My approach has been to study the production of plant hormones by the microsymbiot feeling that by altering the production of these compounds bacteria with improved symbiotic properties, increased competitiveness or increased N-fixation, can be developed.

Jim Hunter



## NITROGEN RESEARCH ACCOMPLISHMENTS AND INTERESTS

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The long term objective of my research program is to improve the biological efficiencies of symbiotic  $N_2$  fixation and nitrate uptake and assimilation in the soybean crop. Pursuit of this objective has involved investigation of specific short term objectives. Effects of 1) source of nitrogen nutrition on form of nitrogen transported from roots to shoots and on ionic balance, 2) host plant genotype, nodule symbiont and phosphorus stress on symbiotic  $N_2$ -fixation efficiency and 3) phosphorus stress on carbon and nitrogen utilization in nitrate-dependent plants have been investigated.

The source of nitrogen supplied to soybean plants was shown to have a profound influence of the nitrogen compounds translocated from roots to shoots in the xylem (8, 9). Ureides, allantoin and allantoic acid comprised 80% of the total nitrogen in xylem sap from plants totally dependent on  $N_2$  fixation, and nitrate and asparagine each comprised 45% of the total nitrogen in sap from plants totally dependent on nitrate assimilation. Relative ureide content of xylem sap was highly correlated with relative contribution of symbiotic  $N_2$  fixation to the nitrogen requirement of the plants (9). This work contributed to the elucidation of a complex nitrogen assimilation pathway in soybean nodules and suggested an alternative method of measuring symbiotic  $N_2$  fixation by field-grown soybean plants.

Two determinate soybean cultivars (Ransom and Davis) were shown to differ in capacities to: 1) fix  $N_2$  during vegetative growth 2) mobilize vegetative tissue nitrogen during reproductive growth and 3) sustain  $N_2$  fixation during reproductive growth (2). However, yield of seed dry mass and protein was the same for both cultivars. These results illustrate that equal yield potential can be achieved by different physiological mechanisms associated with seasonal expression of nitrogen metabolic processes. These cultivars have been used as parents in a breeding program to enhance  $N_2$ -fixation capability of the soybean crop.

Symbiotic  $N_2$ -fixation efficiency of a fast-growing Rhizobium fredii strain, USDA 191 was demonstrated to be inferior to that of Bradyrhizobium japonicum USDA 110 when North American soybean cultivars were used as host plants (3). However significant variation in symbiotic  $N_2$ -fixation efficiencies of five soybean cultivar - USDA 191 symbiosis was observed. This work indicated that it would be inappropriate to introduce R. fredii strains into North American agricultural systems until more efficient symbioses can be developed. The quantitative variation in  $N_2$ -fixation efficiency of different host-strain combinations indicated that  $N_2$ -fixation efficiency of the soybean - R. fredii symbiosis is regulated by more than one host plant gene.

A mannitol-utilizing and highly efficient  $N_2$ -fixing derivative (MN-110) was isolated from a laboratory culture of Bradyrhizobium japonicum USDA 110 and characterized (6, 7). The discovery of such an isolate disproved the hypothesis that mannitol utilization by free-living B. japonicum and absence of  $N_2$ -fixation activity in symbiosis with soybean were causally related. This MN-110 derivative was shown to have 20% greater  $N_2$ -fixation efficiency than a

previously characterized  $N_2$ -fixing derivative, I-110. This research suggests that other naturally-occurring, and highly efficient  $N_2$ -fixing derivatives could be obtained by screening single colony isolates derived from laboratory cultures of commonly used B. japonicum strains. Cultures of this highly efficient MN-110 derivative have been given to a commercial inoculant manufacturer.

The effect of phosphorus stress on efficiencies of symbiotic  $N_2$  fixation and nitrate uptake and assimilation in soybean have been examined (4). When plants were totally dependent on  $N_2$  fixation, the primary effect of phosphorus stress on the nitrogen assimilation pathway was to limit nitrogen input via negative effects on nodule initiation, growth, development and functioning. When plants were dependent on nitrate uptake and assimilation, incorporation of soluble-reduced nitrogen into protein and nucleic acids was the step in the assimilatory pathway that was most sensitive to phosphorus stress. Plants totally dependent on symbiotic  $N_2$  fixation had a higher internal phosphorus requirement for optimal growth and nitrogen accumulation than nitrate-dependent plants. The results suggest that phosphorus fertilization recommendations for soybean may need to be adjusted to take into account the amount of inorganic nitrogen (nitrate) available in the soil.

My current research effort is directed toward elucidating effects of phosphorus stress on 1) distribution of energy between nodule host cells and bacteroids 2) nitrogen assimilation within nodule host cells 3) expression of nitrogenase and nitrogenase reductase genes in nodule bacteroids and 4) whole plant carbon and nitrogen utilization in nitrate-dependent soybean plants. Basic knowledge gained from this research could lead to enhanced efficiency of phosphorus fertilizer use in production of soybean and other leguminous crops.

Superior  $N_2$ -fixing strains of rhizobia and bradyrhizobia have been isolated and characterized under glasshouse or growth chamber conditions. When these strains have been used as inoculants for leguminous crops growing in soils with high indigenous populations of nodulating organisms, they have generally not been successful in nodulating the host plant. Many leguminous crops are produced on soils with high populations of indigenous nodulating strains and some studies indicate that indigenous populations tend to be dominated by strains with low  $N_2$ -fixation efficiency (10). Thus maximal utilization of symbiotic  $N_2$  fixation in agricultural production systems will require successful nodulation of leguminous crops with highly efficient  $N_2$ -fixing strains in soils with high populations of competitive but less efficient  $N_2$ -fixing strains. Solving this problem will require knowledge of how environmental stress factors (high or low soil temperature, low soil moisture, high acidity, high aluminum and low fertility) in specific soil systems influence survival, persistence, nodulating-ability and  $N_2$ -fixing ability of symbiotic microorganisms. Some diversity in tolerance of nodulating organisms to single environmental stress factors in the absence of other soil microorganisms has been observed (5). Tolerances to multiple environmental stress factors in the presence and absence of other soil microorganisms need to be evaluated. Comparative biochemical and genetic studies with strains that exhibit tolerances to specific environmental stress factors should help elucidate tolerance mechanisms. Success in selecting soybean genotypes that restrict nodulation by strains of the 123 serocluster of B. japonicum has been achieved (1). This approach should be expanded with soybean and with other leguminous crops.

## Current Research on the Nitrogen Nutrition of Plants

(Cris title: Formation and Excretion of Ammonia by Nitrogen Fixing Microorganisms and Plants)

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Ammonia (NH<sub>3</sub>) and glutamate (glu) are useful nitrogen (N) sources for the differential asymbiotic growth of Bradyrhizobium japonicum subspecies. We have characterized two distinct subspecies of B. japonicum: (i) USDA 110 type requires glu as N source for growth, produces a galactosyl heteropolysaccharide, and reduces either a small quantity or no acetylene; and (ii) NRRL L-259 type utilizes either glu or NH<sub>3</sub> for growth, produces a 4-methyl-glucuronorhamnan, and reduces large quantities of acetylene.

These N sources were also added to enrichment media in order to obtain tryptophan (trp)-catabolic variants of strain L-259, designated tan. The tan variants were characterized by their tan-orange colored colonies on agar medium containing trp, and were shown to produce large quantities of indolepyruvic and indoleacetic (IAA) acids.

Tan variants, exemplified by tan 4b and 18ac, differ in the way glu and NH<sub>3</sub> are assimilated; their assimilation patterns correspond to the N source used in enrichment cultures. Tan 4b, from a glu<sub>14</sub>-trp enrichment medium, and parental strain L-259 both assimilate <sup>14</sup>C-glu actively and <sup>14</sup>C-methylamine (analogue of NH<sub>3</sub>) passively. Strain 110, which does not grow in a medium where NH<sub>3</sub> is the sole N source, does not even assimilate <sup>14</sup>C-methylamine passively. Tan 18ac, from an NH<sub>3</sub>-trp enrichment medium, is similar in these respects to tan 4b and L-259. However, tan 18ac is unlike tan 4b with respect to trp as sole N source for growth. Tan 18ac grows with trp as sole N source, whereas tan 4b does not. Accordingly, tan 18ac is similar to tryptophan-utilizing Klebsiella aerogenes (tut), described by Paris and Magasanik (J. Bacteriol. 145:257, 1981).

Our study of <sup>14</sup>C-glu and <sup>14</sup>C-methylamine uptake and retention indicates that three characteristic phases of N assimilation by bradyrhizobia are detectable because of slow growth. Phase 1 occurs within a few min and suggests active transport across the cellular membrane barrier. Phase 2, showing net uptake-accumulation of substrates, occurs within 30 to 60 min. Phase 3 occurs after 60 to 120 min; metabolic activity is shown by net accumulation or catabolism of substrates.

The N assimilation patterns are being studied to determine whether stability and efficiency of bradyrhizobial inocula are affected when growth is induced by such different N sources as yeast extract, glu, NH<sub>3</sub>, or trp. Early events (up to 3 weeks) in the symbioses are studied within plastic growth pouches to determine soybean growth, root nodulation and infection sites. Studies with exudate-deficient (leached) seedlings indicate that nodulations by tan variants provide supplemental growth hormone as IAA to plants.

### List of Primary Problems in Research Area

1. Characterizing stability and efficiency of bradyrhizobial inoculants that cause soybean nodulations.
2. Relationship of plant auxin, cytokinins, and growth inhibitors to efficient nodulation.
3. Recognition mechanism of soybean root-cells for specific Bradyrhizobium.
4. Investigation of B. japonicum subspecies and strains specificity and mechanism(s) leading to nodulation (plant-microbial interaction).
5. Investigation of host cultivars appropriate for each bradyrhizobial strain (host range).

### Research Priorities for ARS Concerning Nitrogen Research

Before genetic manipulations of bradyrhizobia and plant host-cells can become practical for improving soybean crops, ARS should provide prior knowledge of the genetic traits which are crucial for symbiosis as well as dinitrogen (N<sub>2</sub>) fixation. We think a proper priority is:

1. Continued studies on the desirable traits inherent with both host and microbe. Intensive physiological study of the underlying mechanisms leading to symbiosis.
2. Initiate novel chemical studies on catalytic/biocatalytic mechanisms that lead to either N<sub>2</sub> reduction directly or energy transfer to facilitate N<sub>2</sub> reductions.
3. Initiate novel biological studies that indicate potentials for improving symbiosis and N<sub>2</sub> fixation. For example, plant cell-bradyrhizobial interactions should be assessed with the awareness that not only N compounds are exchanged but auxins and other growth hormones may also be elicited by the symbiont. Also, the "recognition sites" residing in root-cells and the cognizable compound(s) of Bradyrhizobium should be clearly defined. These are only two examples of fresh approaches to the symbiotic phenomenon, a unique partnership that is of mutual benefit.

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## I. Problem in Agriculture

Biological Nitrogen fixation (BNF) is an energetically expensive process. In some legumes, up to 30% of the total utilizable energy produced by the plant is used for nodule metabolism. Energy for  $N_2$  reduction is generated from plant photosynthate by the microbial symbiont in root nodules. Thus carbohydrate supply and the efficiency of carbohydrate use play a major role in controlling the nitrogen fixation process. Before progress can be made in the improvement of symbiotic nitrogen fixation, the physiological and biochemical mechanisms regulating carbohydrate use, energy production and nitrogenase activity in root nodules must be thoroughly understood.

The microbial symbiont utilizes plant photosynthate and transduces it into energy useful for nitrogen reduction. Any gains in the efficiency of energy conservation by nodule rhizobial bacteroids potentially could be translated into gains in crop yield. Currently, knowledge of energy transduction in the microbe is insufficient to determine if the efficiency can be increased by selection or genetic engineering.

### What we know and need to know

Some microbe-legume symbioses have the ability to recycle the  $H_2$  generated by the nitrogen reduction process. In soybeans, it has been demonstrated that such a symbiosis is more efficient and produces a yield increase. This is attributed to increased reductant availability and/or energy conservation by the microbe. The genetics of the process has been determined and genetic modification of the microbe is possible.

The composition of the energy conserving electron transport chain (ETC) of Bradyrhizobium japonicum bacteroids is phenotypic of the DNA homology group. Current knowledge of energy conservation is insufficient to assess whether energy conservation and  $N_2$  reduction can be increased in the bacteroids. Work is needed to determine which components of the ETC are involved in symbiotic nitrogen fixation. This Laboratory is performing these studies using molecular biological techniques to generate site directed mutations in specific components of the energy conserving electron transport chain of B. japonicum.

The nodule depends on an adequate supply of photosynthate to support nodule metabolism. In the nodule, oxygen is needed by the bacteroid to support nitrogen reduction but its concentration must be carefully controlled since nitrogenase is inactivated by  $O_2$ . One of the mechanism of control of oxygen level in the nodule is by a diffusion barrier which is controlled by the plant. Factors controlling the diffusion barrier must be elucidated before symbiotic performance can be guaranteed.

## II. Problem in Agriculture

In most soils in the U.S., more than two-thirds of the nodules of soybeans are occupied by indigenous native rhizobial strains which are not highly effective in BNF. Establishment of superior rhizobial strains is difficult and consequently inoculation in soils where the legume has been cultivated previously seldom provides a positive benefit to the plant. This lack of response has been attributed to undefined phenotypes which confer superior competitiveness and/or superior saprophytic capabilities in the soil and in the rhizosphere. Current knowledge of the physiological and biochemical factors involved in recognition of the microbe by the plant and the subsequent infection process are insufficient to understand the problem of competition and how to establish introduced strains in agricultural soils.

### What we know and need to know

Superior strains of rhizobia have been developed by selection. This work and further improvement of strains by genetic manipulation should continue.

Many genes, including "common nodulation" genes, "fix" genes and host specificity genes have been identified but the physiological factors involved in host specificity, nodulation and rhizosphere competence are unknown. Work in this area should be encouraged. Several reports have indicated that extracellular polysaccharides (EPS) may be important. This Laboratory is isolating and characterizing EPS mutants to determine their role in symbiosis.

Leon V. Kochian, Plant Physiologist:

We have recently initiated a research program aimed at studying the physiology and biophysics of nitrate absorption across the plasmalemma of root cells. We have constructed  $\text{NO}_3^-$  and  $\text{H}^+$  selective microelectrodes (tip. dia. =  $0.5\mu\text{m}$ ) and have used these microelectrodes to study  $\text{NO}_3^-$  and  $\text{H}^+$  transport at the root surface of noninduced and  $\text{NO}_3^-$ -induced maize seedlings in conjunction with electrophysiological studies of  $\text{NO}_3^-$  uptake. We have found that in induced roots,  $\text{NO}_3^-$  uptake is characterized by a rapid small depolarization of the membrane potential ( $E_m$ ), followed by a slower and larger overall hyperpolarization of  $E_m$ . The depolarization is independent of  $\text{NO}_3^-$  concentration, while the hyperpolarization is concentration-dependent with a  $K_m$  of about  $50\mu\text{M}$ . In noninduced roots, only a slow hyperpolarization of  $E_m$  is seen, in response to  $\text{NO}_3^-$  application. A similar electrical response is seen associated with  $\text{Cl}^-$  uptake in both induced and noninduced roots, but differential responses of  $\text{Cl}^-$  and  $\text{NO}_3^-$  uptake to inhibitors indicates that the two ions are transported by separate systems. Experiments in which  $E_m$  and  $\text{NO}_3^-$  uptake were measured simultaneously demonstrated conclusively that the  $\text{NO}_3^-$ -induced electrical responses are the result of accelerated  $\text{NO}_3^-$  uptake that occurs after exposure of noninduced roots to  $\text{NO}_3^-$ . Further research based on simultaneous measurements of  $E_m$ , and  $\text{NO}_3^-$  and  $\text{H}^+$  transport, was conducted under various conditions (different pH values,  $\text{NO}_3^-$  concentrations, inhibitors, fusicoccin, etc.), has produced results that allow us to speculate (in agreement with some previous researchers) that the initial depolarization is due to  $\text{NO}_3^-$ - $\text{H}^+$  cotransport ( $\text{H}^+:\text{NO}_3^-$  stoichiometry  $> 1$ ), while the subsequent hyperpolarization is either the result of an acceleration of the  $\text{H}^+$  pump, or the operation of a separate, hyperpolarizing  $\text{NO}_3^-$  influx pump.

John F. Thompson, Plant Physiologist:

In the past, our plant studies included the regulation of nitrate reductase assimilation of  $\text{NH}_3$ , and breakdown of urea. We also have done research on amino acid biosynthesis and metabolism.

Leon V. Kochian, Plant Physiologist:

At the cellular level, one significant priority should be the molecular characterization of the transport systems involved in  $\text{NO}_3^-$  and  $\text{NH}_4^+$  uptake into the root. At this time, the molecular mechanisms of absorption of these ions is poorly understood. However, with the recent advances in molecular biology techniques, it may be possible to identify the plasmalemma proteins involved in  $\text{NO}_3^-$  and  $\text{NH}_4^+$  absorption, and to identify, clone, and sequence the genes that encode for the transport proteins. If this can be accomplished, then a number of exciting avenues of research will be opened. First, it should then be possible to obtain significant amounts of pure transport protein which could be used to study the three-dimensional structure of the proteins, in order to learn more about their function, and to reconstitute the proteins into artificial lipid systems to study their transport function. Also, having access to the genes encoding for these transport proteins would allow researchers to study the expression of these genes in the root (and throughout the plant) during various developmental periods and in relation to different growth conditions. Through this type of molecular approach, we may be able to elucidate the mechanisms by which plants acquire nitrogen from the soil, which should allow us to develop breeding and fertilization strategies that lead to more efficient nitrogen nutrition of crop plants.

John F. Thompson, Plant Physiologist:

Understanding is needed to achieve greater nitrate utilization by plants, so that less nitrate gets into the ground water and into runoff water.



May 4, 1989

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Report for the ARS Working  
Conference on Nitrogen Research

Achievements

I. Strain improvement. The discovery of mutant strains with enhanced nodulation and nitrogen fixation (Kuykendall and Hunter, 1988, patent pending) was a useful by-product of molecular biology studies on a nitrogen-fixing microsymbiont of soybean, Bradyrhizobium japonicum. Characterization of amino acid auxotrophs of Bradyrhizobium has also revealed that leucine-requiring mutants are symbiotically competent whereas strains that require proline, tryptophan, or histidine are not (Kummer and Kuykendall, in press).

II. Microbial ecology. A genetically marked B. japonicum strain, I-110 ARS, was first shown over a decade ago to be useful in field studies on the ecological fate of inoculum strains (Kuykendall and Weber 1978) and Kuykendall et al. (1982) demonstrated that a compatible host soybean increased nodule occupancy by an inoculum strain in the subsequent crop. Kuykendall (1989, in press) has recently shown conclusively that long-term persistence in soil of the facultative symbiont is enhanced by soybean nodulation.

III. Genetic analysis. A gene mapping program has been developed to determine linkage relationships among nif and nod genes in the Bradyrhizobium genus. Unlike the situation in fast-growing Rhizobium species, these gene clusters are not closely linked in the slow-growers. Bradyrhizobium DNA homology groups I and II were found to exhibit major phenotypic differences in fatty acid composition and antibiotic resistances (Kuykendall et al. 1988). Genetic linkage analysis of new genetic markers will provide a better understanding of the genetic map of Bradyrhizobium japonicum.

Relevant Publications

1. Kummer, R. and L. D. Kuykendall. 1989. Symbiotic properties of amino acid auxotrophs of Bradyrhizobium japonicum. Soil Biology and Biochemistry (in press).
2. Kuykendall, L. D. 1989. Influence of Glycine max nodulation on the persistence in soil of a genetically marked Bradyrhizobium japonicum strain. Plant and Soil (in press).
3. Kuykendall, L. D., Devine, T. E., and Cregan, P. B. 1982. The positive role of the host plant (soybean, Glycine max) in the establishment of Rhizobium japonicum in subsequent crops of soybeans. Current Microbiol. 7:79-81.

4. Kuykendall, L. D. and Hunter, W. J. 1988. Enhancement of nitrogen fixation with Bradyrhizobium japonicum mutants (patent pending, serial no. 07/325,184).
5. Kuykendall, L. D., Roy, M. A., O'Neill, J. J., and Devine, T. E. 1988. Fatty acids, multiple antibiotic resistance, and DNA homology among Bradyrhizobium japonicum strains. *International Journal of Systematic Bacteriology*. 37:358-361.
6. Kuykendall, L. D. and Weber, D. F. 1978. Genetically marked Rhizobium identifiable as inoculum strain in nodules of soybean plants grown in fields populated with Rhizobium japonicum. *Appl. Environ. Microbiol.* 36:915-919.

### Issues

Nitrogen research within ARS has produced both interesting and practical results (see all the reports submitted for this conference). Sufficiency in crop plants is our main concern. Improved research opportunities are important issues facing ARS in these endeavors:

- (1) Improved communication among groups at different locations in the U.S. is needed. This workshop should help to provide coordination of diverse research efforts.
- (2) Fruitful collaboration should be actively pursued among scientists across locations and disciplines .
- (3) We should continue our emphasis on quality contributions. Adequate financial support must be given to high quality research programs.

### Future research imperatives

- (1) Investigations of Bradyrhizobium genetics need to be intensified in order for practical success in improving nitrogen fixation in soybean to become a reality during our lifetime.
- (2) A focused and interdisciplinary approach to research on competition among rhizobial strains is needed to solve the problem.
- (3) Plant genome and bacterial chromosome mapping should emphasize genes controlling nodulation and symbiotic nitrogen fixation in soybean/Bradyrhizobium.

## USDA-ARS WORKING GROUP ON NITROGEN RESEARCH

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### MAJOR RESEARCH INTERESTS:

Biology of root pathogens and beneficial rhizosphere microbes, especially mycorrhizal fungi and rhizobacteria in relation to nursery and other horticultural crops, biological control, reduction of plant stresses, and nitrogen fixation.

### CURRENT RESEARCH ACTIVITIES PERTINENT TO NITROGEN CONFERENCE:

Biology and control of several nursery pathogens such as *Phytophthora* spp. causing stem and root rot of apple rootstocks. Experiments to determine the potential for enhanced growth and biocontrol using combinations of VA mycorrhizal fungi and bacterial and fungal biocontrol agents.

Interactions between mycorrhizal fungi (VA, ericoid, and ecto-mycorrhizal fungi) with the most promising bacterial and fungal biocontrol agents. Fungal biocontrol agents: Trichoderma, Talaromyces, Gliocladium; Bacterial biocontrol agents: Pseudomonas, Enterobacter, Serratia, Bacillus, Erwinia, Alcaligenes, Streptomyces, and Agrobacterium. Objectives are to determine if any of the biocontrol agents cause any adverse effects on mycorrhizae, and to identify compatible combinations of mycorrhizal fungi and biocontrol agents.

Effects of mycorrhizal fungi on plant responses to soil drought, including mechanisms involved and methods to evaluate and select the most effective fungi. Work with VA mycorrhizal fungi is on pigeon pea inoculated with Rhizobium and evaluated for response to soil drought and enhancement of nitrogen fixation. Seven different mycorrhizal fungi are used in this study.

Evaluation of the mechanisms of enhanced nitrogen fixation when *Rhizobium*-inoculated legumes are inoculated with compatible strains of VA mycorrhizal fungi. These studies focus on the nodulation process since nodulation is enhanced so dramatically by inoculation with mycorrhizal fungi.

Some preliminary experiments evaluating the effects of combinations of VA mycorrhizal fungi and free-living nitrogen fixing bacteria like Azospirillum.

### ACHIEVEMENTS RELATIVE TO NITROGEN NUTRITION OF PLANTS:

Demonstrated the additive effect on growth enhancement of legumes inoculated with Pseudomonas putida, Rhizobium and VA mycorrhizal fungi.

Demonstrated the striking differential growth enhancement response of Rhizobium-inoculated pigeon pea inoculated with different strains or species of VA mycorrhizal fungi. The response was not mediated by P nutrition, but was correlated with enhanced nitrogen fixation due to increased number and size of nodules.

Demonstrated the reduction of plant stress response to soil drought when inoculated with different strains and species of VA mycorrhizal fungi.

Demonstrated the lack of correlation between development of P-absorbing external hyphae of VA mycorrhizal fungi and growth enhancement of legumes.

Demonstrated the shift in rhizosphere populations of bacteria as plants become mycorrhizal, due to morphological and/or physiological changes in the host plant, including changes in root exudation patterns.

## FUTURE PROBLEMS AND NITROGEN RESEARCH PRIORITIES FOR ARS

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### PRIORITY NITROGEN RESEARCH TOPICS: (not in priority order)

1. Nitrogen budgets for a wide range of crops. High levels of nitrogen are showing up in run-off water and are contaminating ground water. For various cropping systems where high levels of nitrogen are being applied, it is imperative to determine where all the nitrogen goes.
2. Develop improved strategies for increased efficiency of plant use of nitrogen fertilizers. Research in this area could emphasize (a) improved ways to retain nitrogen in the root zone, and (b) improved efficiency of nitrogen uptake by roots, such as by mycorrhizal fungi, allowing for reduced rates of application.
3. Increased knowledge of microbial nitrogen fixation systems, including symbiotic and free-living nitrogen fixers. More research is needed to improve the potential for these microbial systems to biologically fix nitrogen, including a detailed examination of the factors most affecting nodulation and survival and colonization of the rhizosphere and endorhizosphere of plant roots by nitrogen fixing microbes.
4. Strategies for inoculating plants with combinations of microorganisms that are known to provide benefit to plant growth and health. Beneficial effects to crop plant growth and health have been demonstrated for nitrogen fixing bacteria, fungal and bacterial biocontrol agents, and mycorrhizal fungi. The technology for inoculating with these organisms is being developed separately, but there is tremendous need and opportunity to combine those separate technologies and inoculate with microbial combinations known to be compatible and that can deliver multiple benefits.
5. Development of slow-release nitrogen technology using organic forms. Inorganic sources of nitrogen release in the ionic form which is or can readily become leachable down the soil profile. Organic forms of nitrogen, or ionic forms complexed with organic materials or organic molecules could be developed to slow the rate of leaching and increase the potential for uptake by plant roots, with or without mycorrhizal fungi.



## USDA-ARS WORKING CONFERENCE

May 23-25, 1989

"Nitrogen Research 1989: Current Advances and Future Priorities"

Charles T. MacKown  
Plant Physiologist  
Tobacco and Forage Research Unit

### I. Research Accomplishments

Nitrogen research goals at our location emphasize the development of fundamental knowledge of the involvement of N in plant (tobacco) processes related to crop productivity and quality. Research personnel working on N related problems include individuals with strengths in chemistry, whole plant physiology, and biochemistry. Consequently, our N research covers a number of different areas including accomplishments related to (1) establishment of the relationship between tobacco-specific nitrosamines and the precursors nitrate and alkaloids, (2) regulation and biochemistry of enzymes involved in primary N and C metabolism, (3) characterization of cultivar physiological traits associated with N use efficiency, and (4) evaluation of nutritional effects on net uptake, assimilation, and partitioning of N.

Rather than address each of these areas, in some of which I have not been directly involved, I will summarize selected accomplishments related to my research program. One aspect of my program is the development of fundamental knowledge useful in the selection and control of nitrate uptake, one of the two components of N use efficiency. Experiments were conducted using a well characterized model root system to determine the physiological changes in the inducible nitrate transport and reduction activities following deprivation of nitrate and initial nitrate exposure.

The experiments demonstrated that (a) endogenous root nitrate minimized a rapid decline or complete loss in both the induced nitrate uptake state and the rate of *in vivo* nitrate assimilation; (b) net efflux of nitrate was strongly correlated with the amount of endogenous nitrate and appeared to best fit a passive flux model; (c) short nitrate exposures ( $\leq 1$  h at 0.25 mM) and low nitrate concentration (0.01 mM for 1 h) were sufficient to cause a 4-fold increase in nitrate uptake rate within 3 h, but development of the accelerated nitrate uptake state was less than the fully induced state; and (d) the rapid development of accelerated nitrate transport following a 1 h pulse with 0.25 mM nitrate was followed by a slow rate of decay in nitrate uptake capacity. Information from these experiments has provided a basis for formulating biochemical studies now in progress to identify membrane polypeptides involved in nitrate transport.

## II. Future ARS N Research Directions

Plant N use efficiency is dictated by processes affecting N acquisition and N utilization for biomass production. For non-legumes, N acquisition is dependent upon absorption of inorganic N sources. In the past 25 years a considerable amount of information on physiological aspects of nitrate and ammonium uptake has been developed. However, biochemical information of inorganic N transport is extremely limited. Only recently a few reports have begun to appear which provide data potentially relevant to the biochemical and molecular aspects of nitrate transport. In view of the benefits associated with decreasing losses of applied N and exploiting available resources efficiently, fundamental research designed to identify, select, and control desirable N use efficiency traits should be a promising approach.

Current ARS policy emphasizes and links plant research at a location to a specific commodity. In the development of fundamental research, ARS policy should encourage creative approaches unrestricted by commodity and industry clientele. This change would insure continued progress in fundamental areas of plant science research benefiting a diverse range of commodity groups.

## Current Research on the Nitrogen Nutrition of Plants

(Cris title: Formation and Excretion of Ammonia by Nitrogen Fixing Microorganisms and Plants)

J. W. Newton, Northern Regional Research Center, ARS-USDA, Peoria, IL 61604

We recently reported (BB Acta, 891, 1987, pp. 49-55; Plant Science 60, 1989, pp. 61-66) that herbicidal inhibitors of photosystem II induce ammonia liberation from both algal and plant cells. The released ammonia can arise from newly-fixed or freshly-assimilated ammonia or, especially in leaves, from ammonia accumulated over several hours. This ammonia liberation occurs at low inhibitor concentrations similar to that which inhibits CO<sub>2</sub> fixation. The effect is enhanced by light, but it can even occur in the dark. Inhibitors of photosystem II are also excellent inhibitors of ammonia uptake by plant and algal cells. From these studies we have postulated that the herbicide-binding 32 kDa (D1) protein of photosystem II may play a regulatory role controlling carbon and nitrogen assimilation in plants. This hypothesis places the regulatory point for carbon and nitrogen metabolism at the very first stage of electron transport rather than a later stage or one involving a competition for carbon skeletons.

Many observations made over several decades of plant research can be explained by this hypothesis, including the effects of ammonia on the Hill reaction and photophosphorylation, observations on the effects of triazines on crop plants and the reduction of herbicidal injury in plants fertilized with ammonia.

These studies suggest that, in addition to providing information on the primary reactions of photosynthesis, studies on photosystem II may provide important information about nitrogen transport in plants. Such studies could lead to plants with improved nitrogen fixation and/or assimilation activity or to plants which excrete ammonia and would be useful as soil reclamation crops or as green manures.

### List of Problems in Research Area

Identification of a relationship between photosystem II and ammonia metabolism suggests that it may be possible to study, *in vitro* and *in vivo*, competition between ammonia and herbicide binding to the D1 protein of plants. Furthermore, genetic modification of the D1 protein could lead to plants with specific desirable traits such as enhanced ammonia excretion or assimilation. Herbicide resistant algae and plants have been identified, but their nitrogen metabolism, to our knowledge, has never been studied directly, and it is possible that interesting properties of these mutants have been overlooked.



## Research Priorities for ARS Nitrogen Research

As with most research, it is hard to predict which areas may be most fruitful in providing basic information of value to agriculture. Consequently, research should proceed along a broad front, to include the following:

1. Work on the chemical mechanism and catalyses of nitrogen reduction and N transformations.
2. Genetic studies on nitrogen fixation, nitrogen metabolism, development of mutants with unique properties.
3. Biochemistry of symbioses, including recognition, factors involved in establishment of the symbiotic state, microbial-root relationships and search for new and unusual symbioses.
4. Transformation of nitrogen in soils including fixation, denitrification, and interaction of N cycle reactions with added nitrogen and cycle inhibitors.
5. Physiology of nitrogen metabolism in plants, algae and nitrogen fixing microorganisms.

William E. Newton  
Albany, CA

## I. CURRENT RESEARCH

A. Organization & Functioning of Nitrogenase. To assist in determining the mechanism of biological N<sub>2</sub> fixation, we have been involved in sequencing (with D. Dean, VA Tech) the A. vinelandii nif gene cluster. In particular, using the deduced amino acid sequences in comparison with others already (and since) published, we have targeted specific residues as likely prosthetic group-binding sites and have introduced specific, single substitutions via a site-directed and gene replacement strategy. As the MoFe protein component contains the N<sub>2</sub>-binding site, it has been our target. All 8 strictly conserved cys residues have been replaced with ser and ala individually, 3 of the conserved his have been substituted by asn, and 2 conserved gln have been substituted by glu, lys, or asn. We have determined the EPR-spectroscopic and catalytic sequences of these disruptions and find normal, no, or slow growth on N<sub>2</sub>, plus altered substrate discrimination. Kinetic and spectroscopic analyses suggest that the binding domains of both prosthetic group types have been disturbed.

B. Biosynthesis of Iron-Molybdenum Cofactor. As the nifDK products (the MoFe protein subunits) are not required for FeMo-co biosynthesis, FeMo-co is likely synthesized elsewhere and then inserted into the apoprotein. We (with Dean, VA Tech) have sequenced the nifEN genes, whose products are necessary for FeMo-co biosynthesis-maturation, and find striking homology with nifD and nifK, respectively. These results indicate a structural and functional relationship between the nifEN protein and the MoFe protein, and suggest that the nifEN protein forms a template on which FeMo-co is built before donation to the apo-MoFe protein.

C. Structure and Function of Isolated FeMo-co. FeMo-co is the active site of the MoFe protein. By a combination of x-ray absorption (XAS), Mossbauer and EPR spectroscopies, plus electrochemistry, applied to FeMo-co after purification by gel filtration, we found that:

i) FeMo-co can exist in 3 oxidation states, oxidized (ox), semireduced (s-r), and reduced (red) with the ox to s-r conversion being quasi-reversible and involving 1 electron which is not accommodated on the Mo atom;

ii) its Mo has an altered nearest neighbor arrangement MoO<sub>3</sub>S<sub>3</sub>Fe<sub>3</sub>, compared with its protein matrix, MoO<sub>2</sub>S<sub>4</sub>Fe;

iii) the addition of 1 thiolate per Mo alters this arrangement to  $\text{MoO}_3\text{S}_4\text{Fe}_3$ , sharpens its EPR signal and makes the ox to s-r change electrochemically reversible;

iv) FeMo-co may exist in a variety of forms depending on oxidation state and the solvent acid/base status;

v) FeMo-co is capable of the catalytic reduction of acetylene to ethylene, and

vi) FeMo-co appears to be a symmetrical entity.

Our goal is to determine how  $\text{N}_2$  is fixed biologically such that targets and methodologies can be generated for beneficial modifications to be made to nitrogenase by genetical techniques for agricultural benefit.

## II. PRIMARY ISSUES FACING ARS.

A. How does nitrogenase catalyze the reduction of  $\text{N}_2$  under ambient conditions? Can it be manipulated beneficially? How is it regulated?

B. Why are 20-odd genes required? What do they do?

C. What is the role(s) of the alternative system(s)? Do they occur in Rhizobium?

D. Can a "nif gene package" be assembled and transferred stably to other bacterial species/genera? To plants? Where?

E. Do other agriculturally useful, symbiotic,  $\text{N}_2$ -fixing systems exist?

## III. RESEARCH IMPERATIVES.

A. Characterize all nif gene products, their role in  $\text{N}_2$  fixation, and how to manipulate them for agricultural benefit.

B. Determine the organization and structure of the nitrogenase component proteins to overcome current inefficiencies and limitations (e.g.,  $\text{H}_2$  evolution, specific ATP requirement, specific Fe protein as reductant).

C. Determine the existence in Rhizobium and characterize the role(s) of the alternative  $\text{N}_2$ -fixing systems.

D. Integrate these efforts ARS-wide.

T. W. Ruffy, Jr.  
Crops Research Lab., Oxford, NC  
Dept. of Crop Sci., N. C. State Univ.

The research program is directed towards understanding regulation of inorganic nitrogen assimilation by crop plants. The long-term goal is to modify plants genetically and improve nitrogen use efficiency. Current research is centered on mechanisms controlling the assimilation of nitrate in vivo, utilizing experimentation with  $^{13}\text{N}$  and  $^{15}\text{N}$  isotopes.

1. Achievements. Results indicate that there are primarily three rate-limiting processes in the nitrate assimilation pathway leading to protein synthesis; the processes appear to control assimilation on a daily basis and they are the main response points when environmental or nutritional stresses are imposed. The processes are 1) uptake of nitrate into the root from the rhizosphere, 2) transport of nitrate out of the root into the xylem, and 3) nitrate reduction in leaves. The processes appear to be the major restriction points when plants have a limited energy supply, when plants are exposed to low temperatures, excessive acidity or excessive Al in the rhizosphere, and when plants are experiencing phosphorus and potassium deficiency. The two transport processes are generally more important, quantitatively. We have not been able to find a stress condition which disrupts the nitrate assimilation pathway beyond  $\text{NO}_3^-$  reduction in the protein synthesis reaction sequence.

2. Primary Issues. It is known with a reasonable degree of certainty that nitrate fertilizer is a large expense for farmers and that nitrate from applied fertilizer is a significant contributor to deterioration of water supplies, and thus, that research efforts to improve nitrate use efficiency must proceed. Breeding programs have established that genetic variability exists for nitrate use efficiency. Therefore, direct molecular manipulations of specific control processes to improve efficiency may be possible with time. The primary issue in achieving that goal is whether ARS can assemble the necessary research teams and provide them with adequate resources. An integrative effort is imperative, involving plant breeding, physiology, biochemistry, and molecular biology. Relevant research issues are pointed to in the following list of research imperatives.

3. Future Research Imperatives. Plant breeding programs need to be instituted on a large scale to begin selecting for increased nitrate use efficiency. In a parallel pursuit of plant manipulation using genetic engineering, physiologists and biochemists need to:

- a. Isolate and characterize the protein(s) involved in nitrate transport into root cells and determine the feedback controls governing its activity.
- b. Define the transport system regulating nitrate release from the root symplasm at xylem parenchyma cells, which seems to be involved in the feedback control of uptake and to be the cellular receptor site for signals originating the shoot.
- c. Determine the physiological basis for root proliferation in zones of high fertility and for sustaining root growth during reproductive development of the plant.

d. Investigate the relationship between delivery of reduced nitrogen to leaf growth sinks and the associated "feedback" effects on photosynthesis and carbohydrate formation.

Thomas R. Sinclair

Plant Stress and Protection Unit

Gainesville, FL

Activities

- (1) Regulation of Nitrogen Fixation Rates in Legumes. It was recognized more than a decade ago that direct regulation of symbiotic nitrogen fixation rates may well be associated with oxygen flux into nodules. We have completed a number of studies to quantitate the permeability of the oxygen diffusion barrier in nodules. It was found this barrier is significant in regulating the fixation rates of individual nodules. We observed both in laboratory and field experiments that soybean nitrogen fixation rates and nodule respiration rates are tightly regulated by dynamic adjustments in the diffusion barrier. The permeability of the barrier adjusts in response to changes in rhizosphere oxygen concentration, but barrier permeability is not responsive to ammonia gas. Under well-watered conditions, permeability is closely associated with soil temperature.
- (2) Drought Limitation on Symbiotic Fixation. Early studies on symbiotic nitrogen fixation showed the process was very sensitive to soil dehydration. Both greenhouse and field experiments have shown fixation to be the first physiological process to respond to soil dehydration. Even modest declines in available soil water can adversely effect nitrogen fixation rates. We have identified in preliminary studies variation among genotypes in sensitivity of nitrogen fixation to soil drying. Field and laboratory studies are underway to confirm this variation and identify the physiological basis for such differences.
- (3) Regulation of Nodulation by Host Genotypes. The southern germplasm was screened for variation in number and mass of nodules at Stage R3. Considerable variation among genotypes was found with large differences existing for both nodulation number and mass. The differences in nodulation traits were found to be consistent across environments. Genetic studies have shown these traits to be heritable. Currently, advanced lines are being studied to examine the genetic association between seed yield and greater nodulation.
- (4) Incorporation of Nitrogen Effects in Crop Yield Simulations. We had developed simple, mechanistic models to simulate crop growth as a function of solar radiation, temperature, and precipitation. These models have been found to work well for grain legumes, maize, and wheat. The next step is to incorporate nitrogen response functions into the models. The greatest ambiguities in incorporating nitrogen into the model include nitrogen supply rate from the

rhizosphere and the allocation of nitrogen to leaves. The allocation of nitrogen to leaves is particularly important because leaf area growth is dependent on nitrogen availability and because crop radiation use efficiency is dependent of leaf nitrogen content.

### Future Research Imperatives

- (1) Regulation of Nitrogen Fixation Rates in Legumes. Considerable information regarding the basic processes regulating nitrogen fixation rates yet needs to be resolved. A basic understanding of the processes which regulate nitrogen fixation are required before hypotheses for improving rates can be formulated. The basic mechanisms which cause the permeability of the diffusion barrier to adjust are unknown. Information on the response of the diffusion barrier to various compounds including nitrate needs to be resolved. Identification of the key factors regulating the diffusion barrier may provide a means for adjusting nitrogen fixation rates to the benefit of the crop.
- (2) Drought Limitation on Symbiotic Fixation. Drought is usually the most serious environmental stress imposed upon crops. Legumes are, therefore, especially vulnerable because of the loss of nitrogen fixation rates very early in the soil dehydration cycle. For forage legumes, as well as grain legumes, performance may be significantly enhanced if nitrogen fixation was made less sensitive to drought. Important work is required to understand the physiology of nodule drought sensitivity and to evaluate the potential for crop improvement.
- (3) Incorporation of Nitrogen Effects in Crop Yield Simulations. Other than water stress, inadequate nitrogen is probably the most common limitation to crop yield. In the current era of competing concerns about crop yield, economically efficient fertilizer use, and environmental degradation, simulation models may provide important tools for examining these problems. Nevertheless, key physiological understanding of nitrogen availability in the plant and its use to produce yield are not known. Important studies of whole plant physiology are required to understand the basic processes and develop realistic, mechanistic response functions. Regulation of nitrogen uptake from the rhizosphere and the use of nitrogen in leaf growth remain key physiological activities which need resolution for fully understanding crop performance in environments with limited nitrogen availability.

## CONTRIBUTION TO "NITROGEN NUTRITION OF PLANTS" WORKING GROUP

C.P. VANCE, ST. PAUL, MN

May 22-25, 1989

### Current Research Objectives and Achievements

1. Assimilation of fixed N by legume root nodules. Plant enzymes associated with the assimilation of ammonia derived from  $N_2$  fixation are crucial to nodule function. Many of these same enzymes are also required for the assimilation of N derived from  $NO_3^-$ . Over the last four years we have purified and characterized root nodule glutamate synthase (GOGAT), aspartate aminotransferase (AAT), and phosphoenolpyruvate carboxylase (PEPC). More recently we have produced antibodies to these proteins in order to evaluate the expression of polypeptides comprising the native enzymes.
2. Carbon Nutrition for  $N_2$  fixation. Nonphotosynthetic  $CO_2$  fixation via root nodule phosphoenolpyruvate carboxylase (PEPC) has been implicated in providing carbon for energy in  $N_2$  fixation and for skeletons for N assimilation. We have demonstrated that nonphotosynthetic  $CO_2$  fixation can provide 25 to 35% of the carbon required to N fixation and N assimilation. Carbon fixed by PEPC is directly incorporated into asparagine and aspartate which are the major N transport products in many legume species. Carbon fixed by PEPC is also incorporated into TCA cycle organic acids, primarily malate and succinate. These organic acids are used by  $N_2$  fixing bacteroids for energy to fuel nitrogenase. When wild type nodules are fed  $^{14}CO_2$  bacteroids rapidly accumulate radioactive organic acids. However, when nodules formed by dicarboxylic acid mutants are fed  $^{14}CO_2$  no radioactivity accumulates in the bacteroids.
3. Plant Gene Expression During Nodulation. Legume root nodule formation is a complex developmental sequence requiring the interaction of two genomes, the host plant and Rhizobium bacteria. Using in vitro translation of polyA<sup>+</sup> RNA and 2-D electrophoresis we have identified at least 25 plant gene products that are induced and/or enhanced during root nodule development. Northern blotting and western analysis have been used to characterize transcription and translation of six of these nodule specific gene products.
4. Plants Mutants for Nodulation and  $N_2$  Fixation. Genetic evaluation of plant traits requires phenotypic and genotypic markers. Such markers for legume  $N_2$  fixation are the presence or absence of nodules and the loss of function of root nodules. Our multidisciplinary group at St. Paul has identified, characterized and released alfalfa germplasm having no nodules and having nodules which are ineffective. To date we have identified approximately seven genes in alfalfa that are required for effective nodule formation. We are currently attempting to generate similar mutants in Lotus and Lupinus. We are also collaborating with other groups on characterizing mutants in Pisum and Vicia.



5. Selection for Enhanced N<sub>2</sub> Fixation. For the past 13 years we have been involved in a multidisciplinary effort to select plants having traits associated with enhanced N<sub>2</sub> fixation. During this period we have selected for various morphological and biochemical traits including: 1) nitrogenase activity; 2) nodule number and mass; 3) root and shoot mass; 4) root nitrogen content; 5) shoot nitrate reductase; 6) nodule soluble protein; and 7) nodule nitrogen and carbon assimilating enzyme activity.

#### Primary Issues Facing ARS

1. Delivery of a useful product to public either as germplasm or inoculum.
2. Identification of plant and rhizobial genes controlling nodulation and competitiveness and manipulation of these genes for improved N<sub>2</sub> fixation.
3. Addressing how legumes perform in high temperatures, reduced water, increased CO<sub>2</sub> conditions.
4. What are the agriculturally important genes contributing to improved quality, production, persistence?
5. Is mutation breeding/selection an effective avenue for understanding the genetics of plant traits.
6. What is the role and importance of N<sub>2</sub> fixation via legumes to companion crops and/or fellow crops.

#### Research Imperatives

1. Identify plant genes of agricultural importance, characterize their products, map these genes, and determine how they can best be manipulated to improve plant performance and quality.
2. Identify rhizobial genes controlling competitiveness and introduce such genes into superior Rhizobium strains.
3. Evaluate the importance and economic benefits of increased use of legumes in sustainable agriculture.
4. Continue with or enhance efforts to generate plant mutants for nodulation and N<sub>2</sub> fixation and use such mutants in isolating the plant genes controlling N<sub>2</sub> fixation.
5. Continue fundamental studies of N<sub>2</sub> fixation, N-assimilation, C-assimilation and host-microbe interactions to expand our knowledge base.

REPORT TO THE ARS WORKING CONFERENCE ON NITROGEN RESEARCH  
by Sara Wright  
Appalachian Soil and Water Conservation Research Laboratory  
Beckley, WV 25802-0867

Achievements as related to nitrogen nutrition of plants:

The work done at the ASWCRL on nitrogen nutrition of plants has involved the enhancement of symbiotic nitrogen fixation and the contribution of vesicular-arbuscular mycorrhizal fungi to the enhancement of nitrogen fixation. The use of both plant symbiotic associations is for low-input forage systems on hill land soils of Appalachia. Many soils in this area are acidic, low in phosphorus, and contain high level of phytotoxic aluminum.

Benchmark soils were chosen which represented large areas of potentially productive use for forages. Greenhouse studies determined the general chemical limitations to nodulation by rhizobia in these soils. The current limitation to precise definition of soils with chemical toxicity to symbiotic nitrogen fixation is the inability to accurately determine phytotoxic Al. Further work using soils must await development of more accurate chemical analytical methods.

Simultaneously, new methods to trace introduced rhizobia and mycorrhizal fungi in soil were developed. Immunological methods using monoclonal antibodies were developed to identify strains of rhizobia and species of mycorrhizal fungi. These immunological tests have been used to define soil chemical properties influencing competition among strains of rhizobia, and to identify colonization by a specific mycorrhizal fungus on roots.

Primary issues facing ARS in this area of endeavour:

The primary issue facing ARS is utilization of resources. Solving problems with interdisciplinary groups of scientists would be the best use of money and manpower. Scientists who can work together in a compatible fashion, but are located in several different ARS laboratories, should identify goals and make group efforts to solve the problem. CRIS's written for this type of cooperative research should be encouraged.

Expand funding in Competitive Grants to overcome inequities in funding of research.

For two-year post-doctoral fellowship grants awarded to ARS scientist, require more comprehensive proposals and review these more thoroughly.

Future research imperatives (over the next 5 years):

Define taxonomic relationships among VAM fungi and develop easy identification methods to determine the role of individual species in plant nutrition, particularly legumes.

Enhance current symbiotic systems by determining factors influencing root infection and unstable performance of BNF.

Support training in practical aspects of legume-Rhizobium technology.



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