PD-AAT-414

AGENCY FOR INTERNATIONAL DEVELOPMENT

PROJECT DATA SHEET

1. TRANSACTION CODE: A (A = Add, C = Change, D = Delete) — Amendment Number

DOCUMENT CODE: 3

2. COUNTRY/ENTITY: Bureau for Science and Technology

3. PROJECT NUMBER: 936-4177

4. BUREAU/OFFICE: Office of Agriculture — 10

5. PROJECT TITLE (maximum 40 characters): Improved BNF thru Biotech

6. PROJECT ASSISTANCE COMPLETION DATE (PACD): MM 09 DD 30 YY 96

7. ESTIMATED DATE OF OBLIGATION (Under 'B.' below, enter 1, 2, 3, or 4)
A. Initial FY 86 B. Quarter 2 C. Final FY 95

8. COSTS ($000 OR EQUIVALENT $1 =)

A. FUNDING SOURCE	FIRST FY 86 B. FX	C. L/C	D. Total	LIFE OF PROJECT E. FX	F. L/C	G. Total
AID Appropriated Total	$200		$200	$19,990		$19,990
(Grant) S&T/AGR	(200)	( )	(200)	(9,690)	( )	(9,690)
(Loan)	( )	( )	( )	( )	( )	( )
Other AID 1. Missions, Regional						
Other AID 2. Bureaus & AID/W	-0-		-0-	10,300		$10,300
Host Country						
Other Donor(s)						
TOTALS	$200		$200	$19,990		$19,990

9. SCHEDULE OF AID FUNDING ($000)

A. APPROPRIATION	B. PRIMARY PURPOSE CODE	C. PRIMARY TECH. CODE 1. Grant	2. Loan	D. OBLIGATIONS TO DATE 1. Grant	2. Loan	E. AMOUNT APPROVED THIS ACTION 1. Grant	2. Loan	F. LIFE OF PROJECT 1. Grant	2. Loan
(1) FN	101 I	011		-0-		9,690		9,690	
(2)									
(3)									
(4)									
TOTALS				-0-		9,690		9,690	

10. SECONDARY TECHNICAL CODES (maximum 6 codes of 3 positions each): 000 | 070 | 080 | 953 | 963 | 968

11. SECONDARY PURPOSE CODE: 129

12. SPECIAL CONCERNS CODES (maximum 7 codes of 4 positions each)

A. Code	BF	R/AG	TECH	XII			
B. Amount	$1,955	$9,476	$13,642	$15,834			

13. PROJECT PURPOSE (maximum 480 characters)

The purpose of this project is to: 1) increase the efficiency of nitrogen fixing microorganisms adapted to LDC conditions through methods of biotechnology; 2) promote the use of BNF in LDCs by assisting them to adapt, use, and disseminate information about BNF; and 3) increase their capacity to produce and distribute BNF inocula.

14. SCHEDULED EVALUATIONS
Interim MM 11 YY 88 MM 11 YY 91 Final MM 11 YY 96

15. SOURCE/ORIGIN OF GOODS AND SERVICES
☒ 000 ☐ 941 ☐ Local ☐ Other (Specify)

16. AMENDMENTS/NATURE OF CHANGE PROPOSED (This is page 1 of a ____ page PP Amendment.)

17. APPROVED BY
Signature: Anson R. Bertrand
Dr. Anson R. Bertrand
Title: S&T/FA
Date Signed: MM 10 DD MAY YY 1986

18. DATE DOCUMENT RECEIVED IN AID/W, OR FOR AID/W DOCUMENTS, DATE OF DISTRIBUTION
MM DD YY

30 MAY 1986

ACTION MEMORANDUM TO THE AGENCY DIRECTOR FOR FOOD AND AGRICULTURE, BUREAU FOR SCIENCE AND TECHNOLOGY

FROM: Tejpal Gill, Acting Office Director, S&T/AGR

SUBJECT: Improved BNF thru Biotechnology

Action: Your approval is required to authorize a ten-year project, Improved BNF thru Biotechnology (936-4177) which has an authorized life of project cost of $9,690,000 from the Agriculture, Rural Development and Nutrition Account, Section 103 of the Foreign Assistance Act of 1961, as amended. The total project cost is $19,990,000, of which $9,690,000 will be funded by S&T/AGR under this authorization and $10,300,000 by the regional bureaus, missions, and other AID/W offices under their respective project authorizations.

Discussion: This new project is consistent with the objectives of AID's Food and Agricultural Policy and Strategy to enable LDCs to become self-reliant in food, assure food security, and contribute to broadly-based economic growth. One clear way to achieve these objectives is to increase agricultural production significantly over the next decade using cost efficient methods.

Nitrogen rich soils can increase agricultural production and if nitrogen and other essential nutrients are removed from the soil they must be replenished, in one way or other, to ensure sustained agricultural productivity. This can be accomplished through the addition of chemical fertilizers, which are an economic drain on LDC farmers. Alternatively, there exists a certain class of microorganisms (rhizobia) which can fix free nitrogen in the soil, form mutually beneficial relationships (symbiotic) with leguminous plants, and provide plants the required nitrogen in a useful form. This process is known as biological nitrogen fixation (BNF). An increased use, understanding and improvement in BNF systems benefit the LDCs by increasing crop production and reducing the dependency on expensive fertilizers.

In addition, this project is consistent with S&T's role as defined in the proposed CPSS to: 1) support agricultural research and technology development; 2) provide technical guidance and support to AID missions in the collaborative formulation and implementation of agricultural development projects; and 3) provide on-going linkages for AID and cooperating countries with scientists in U.S. and LDC agricultural institutions and international research centers.

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Project activities will draw on the expertise of scientists from the University of Hawaii to maximize the amount of nitrogen gained from symbiotic nitrogen fixing systems by: 1) performing basic and applied research designed to improve and monitor the rhizobia/plant interaction in tropical environments; and 2) collecting BNF data in different environments that are used to adapt the technologies to LDC environments. The research work will include the identification and manipulation of genes which are responsible for an effective BNF system; and studies to improve the ability of microorganisms (rhizobia) to survive in the tropical soils, adapt to environmental conditions, and nodulate plant roots.

Project activities will provide technical guidance and support to AID missions and other AID entities in the collaborative formulation and implementation of BNF. Collaborative research with LDCs, regional and international research institutions will develop technologies in BNF appropriate for LDC farmers.

A very important step in the BNF system is the production and delivery of the rhizobia to LDC farmers. The rhizobia must be produced in the LDCs and delivered to the farmer in a useful form - the inoculant. This production and delivery step can be carried out as a business enterprise by LDC entrepreneurs. This project will provide training and technical information to encourage developing country entrepreneurs to establish small businesses that produce high quality inoculants.

The project consists of the following five integrated programs or activities which will be supported by S&T/AGR, the University of Hawaii, missions, LDCs, bureaus and other AID/W offices.

Program 1	Genetic technologies for improvement of rhizobium/legume symbiosis for crops and trees;
Program 2	Development of methodologies for monitoring microorganism/plant interaction;
Program 3	Environmental data collection to maximize performance of biological nitrogen fixation;
Program 4	Regional resource centers; and
Program 5	Commercial inoculant technical assistance.

The five programs together form an integrated approach for optimizing the impact of BNF technologies. The improved rhizobia produced under Program 1 will be field tested in Program 3 using the methods developed in Program 2. This testing will be supervised in part by the appropriate regional resource centers to be funded by the regional bureaus and missions under Program 4. Implementation of the technology (production and use) will be facilitated by small LDC enterprises assisted under program 5.

This project is a follow-on to the N-Fixation - Symbiotic project (936-0613) which was established by A.I.D. in 1975 under a contract with the University of Hawaii, and included research, technology transfer, training and networking. The research focussed on the selection and characterization of rhizobia to satisfy the nitrogen needs of plants.

The past research concentrated on identifying the best suited rhizobia for a given legume under specific conditions. This approach is termed improvement by selection and testing. For example, in field trials in 12 country sites, inoculated legumes showed an average yield increase of 24 percent. Under the previous N-Fixation - Symbiotic project, the UH/CTAHR acquired a comprehensive collection of rhizobia specifically adapted to tropical conditions. These strains are characterized and available for distribution to researchers throughout the world. This research, in addition to training, field testing, transferring technology, and networking provided the foundation for the new project.

The new project will build on the established foundation to advance the understanding and control of BNF. It will do this by using traditional methods, as well as the new methods of biotechnology. These new methods will allow problems (e.g., effectiveness of nitrogen fixation, of persistence in the soil, and of greater competitiveness against ineffective soil rhizobia) to be approached at a molecular level. Through genetic engineering nitrogenase genes or symbiotic genes can be manipulated leading to the development of improved microorganisms. Additionally, methods for monitoring the bacteria/plant interaction will be more accurately defined. The fate of the nitrogen-fixing microorganisms (rhizobia) added as an inoculant to the seed or soil will be studied. Such information is needed to improve inoculation methods. This project also plans to adapt and transfer these technologies by providing support to regional centers and involving the private sector in the production and distribution of rhizobia inoculant.

A panel consisting of scientists and an expert in private sector investments will be established to provide advice to the S&T/AGR project manager. The panel will consist of representatives from other S&T/AGR biotech projects (e.g., Biotechnology - Limiting Factors, and Biotechnology - Tissue Culture), a regional BNF resource center (e.g., Thailand), an international organization (e.g., ICRISAT, CIAT), and a representative from the Bureau of Private Enterprise.

Agency Policy: The project will be implemented in accordance with relevant Agency policies, including those on food and agriculture, development of human resource and institutional capabilities, and expanding the role of LDC scientists and institutions.

Funding: This project was originally included in the Project Identification Document (PID) of the umbrella project "Improved Plant and Animal Tolerance to Stress" and was approved by the Senior Assistant Administrator for Science and Technology on March 20, 1986. It was decided that an umbrella approach was not appropriate at that time and that the development of the Improved BNF thru Biotech initiative should proceed as a separate project. S&T/AGR has identified this project as being of high priority, and the funding is included in the S&T/AGR budget plans. The cost estimates are appropriate for the planned level of effort described in the project paper (PP). The FY 1986 OYB has been revised to include $200,000 to initiate this new project.

Justification to Congress: An Advice of Program Change is being cleared by the appropriate AID officials and will be sent to the Congressional Committees shortly.

Clearances Obtained: The Project Paper (PP) for this ten-year project was prepared in consultation with the members of Subcommittee for Biological Nitrogen Fixation of the Sector Council for Agriculture. It was endorsed by the Sector Council on May 6, 1986 with the recommendation that more emphasis be placed on involving the private sector. We have revised the project paper accordingly. (Project Endorsement Sheet and the minutes of the May 6, 1986 Sector Council for Agriculture meeting are attached as Tab C.)

Certification of the Procurement Plan: Certification that the procurement plan for this project paper was developed with full consideration of maximum involvement by minority and women-owned firms, historically Black colleges and universities and minority controlled PVOs in the provision of goods and services, and that the Project is not appropriate for Minority or Gray amendment contracting is attached as Tab A.

Method of Implementation - During the project design stage, it became apparent that a cooperative agreement and a companion basic ordering agreement with the College of Tropical Agriculture and Human Resources (CTAHR), University of Hawaii (UH) is the most efficient and economical method to successfully implement the scope of work outlined in the attached project paper (Tab D). These agreements will be signed in FY 1986 and cover a five-year period. The cooperative agreement will provide the assistance to UH/CTAHR to: 1) strengthen its resource base in biological nitrogen fixation developed over the past decade of cooperation with the Agency and other donors; 2) expand the level and range of its collaboration with U.S., LDC, regional and international institutions; 3) provide assistance for establishing and implementing BNF activities in the LDCs; and 4) provide BNF biotechnology graduate training at the M.S. and Ph.D. levels at UH/CTAHR.

The delivery orders issued under the basic ordering agreement will provide missions, regional bureaus and AID/W offices with short to medium-term technical advisory services for: 1) planning, designing and evaluating programs and projects concerned with BNF; 2) in-country and regional training programs; and 3) testing research results in LDC environments. The practical experiences and on-site information and insights gained during these activities under the delivery orders under the basic ordering agreement will be fed directly back into UH/CTAHR's research, training programs, academic curricula, informational development activities and the applied research agenda developed and implemented under the cooperative agreement. It is also intended that the need and opportunity for delivery orders will be identified by the cooperator in its work under the CA and proposed to the Office of Agriculture, regional bureaus and missions for approval and funding. In addition, UH/CTAHR may receive requests for assistance directly from the Office of Agriculture, regional bureaus, other AID/W offices, missions and/or LDCs.

In consideration of the above and in accordance with Dr. Brady's Delegation of Authority dated December 18, 1981 to the Agency Directors regarding "Predominant Capability Determination"

you have made the determination that UH/CTAHR has predominant capability in BNF tropical agriculture and have signed the required memorandum to M/SER/OP requesting that the cooperative agreement be awarded to UH/CTAHR without competition. (Attached as Tab E). In addition, attached as Tab F is a memorandum to M/SER/OP requesting that the companion basic ordering agreement (BOA), which must "feed back" information and relate directly to the CA activities, be awarded to UH/CTAHR without competition and that the resultant delivery orders under the BOA need not be individually competed. We have consulted with M/SER/OP/W and expect that this request will be approved.

Three comprehensive evaluations are planned over the life of the ten-year project by a panel comprised of scientists, international experts, and a representative from the Bureau for Private Enterprise to determine if the project inputs are being provided as planned, that conditions and covenants of project workscope are being met, and that project outputs are being accomplished as planned. Management reviews will be conducted annually by the S&T/AGR project manager.

Recommendation:

1. That you sign the attached Project Authorization (Tab B) approving $9,690,000 of S&T funds for the ten-year, Improved BNF thru Biotechnology project. Up to $10,300,000 of additional regional bureau, missions and central bureau funds may be provided through contractual instruments during the life of the project.

2. That you sign the Project Data Sheet (Tab B).

Attachment:

Tab A - Certification of the Procurement Plan
Tab B - Project Data Sheet
Tab C - Project Endorsement Sheet and minutes of the Sector Council for Agriculture review
Tab D - Project Paper
Tab E - Memo to M/SER/OP requesting that CA be awarded to UH/CTAHR without competition.
Tab F - Memo to M/SER/OP requesting that the BOA be awarded to UH/CTAHR without competition and that the individual delivery orders under the BOA need not be individually competed.

Clearance:

S&T/AGR,	Tejpal Gill	28Gill	Date	5/28/86
	Lloyd Frederick	[illegible]	Date	5/28/86
	Frances Li	draft	Date	5/16/86
S&T/PO	Gerald F. Gower	[illegible]	Date	29 May 86
GC	Stephen Tisa	[illegible]	Date	
SER/OP,	Joyce Frame	(Draft)	Date	5/27/86

Drafted:LFrederick:MEM:5/1/86:Revised:5/15/86:Revised 5/28/86:BNF4:WANG 3992g

PROJECT AUTHORIZATION

Country: Interregional

Project: Improved BNF thru Biotechnology

Project No. 936-4177

1. Pursuant to section 103 of the Foreign Assistance Act of 1961, as amended, I hereby authorize the project, Improved BNF thru Biotech, involving centrally funded planned obligations not to exceed $9,690,000 in grant funds over a ten-year period from the date of authorization, subject to the availability of funds in accordance with the A.I.D. OYB/allotment process, to help in financing foreign exchange and local currency costs for the project. The project may also include such additional funding up to $10,300,000 contributed for this purpose by regional bureaus, AID missions and AID/W offices, other than S&T/AGR.

2. The purpose of this project is to: 1) increase the efficiency of nitrogen fixing microorganisms adapted to LDC conditions through methods of biotechnology; 2) promote the use of BNF in LDCs by assisting them to adapt, use, and disseminate information about BNF; and 3) increase their capacity to produce and distribute BNF inoculants.

3. The agreements which may be negotiated and executed by the officer to whom such authority is delegated in accordance with A.I.D regulations and Delegations of Authority shall be subject to the following terms and conditions, together with such other terms and conditions as A.I.D. may deem appropriate.

4. Source and Origin of Commodities, Nationality of Services

a. Commodities financed by A.I.D. under the project shall have their source and origin in the cooperating country* or the United States, except as A.I.D. may otherwise agree in writing. Except for ocean shipping, the suppliers of commodities or services shall have the cooperating country or the United States as their place of nationality, except as A.I.D. may otherwise agree in writing.

b. Ocean shipping financed by A.I.D. under the project shall, except as A.I.D. may otherwise agree in writing, be financed only on flag vessels of the United States.

*Each country where research, training, technical, or other assistance takes place under the project shall be deemed to be a cooperating country for the purpose of permitting local cost financing of goods and services for the activity being conducted in such country. Such activities may be undertaken in any country included in A.I.D. geographic code 935.

Anson R. Bertrand
Agency Director
for Food and Agriculture
Bureau for Science and Technology

Clearances:

S&T/AGR,	Tejpal Gill		date 5/27/86
	Frances Li		date 5/16/86
	Lloyd Frederick		date 27 May 86
S&T/PO,	Gerald F. Gower		date 29 May 86
GC	Stephen Tisa		date

Drafted by:LFrederick:MEM:5/7/86:x525-3840 BNF4

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Tab A

Improved BNF thru Biotechnology

Certification of the Procurement Plan

I certify that the procurement plan for this Project Paper (936-4177) was developed with full consideration of maximum involvement by minority and women-owned firms, historically Black colleges and universities and minority controlled PVOs in the provision of goods and services, and that the Project is not appropriate for minority or Gray amendment contracting. We know of no minority institutions with an on-going program of the type required, or the requisite faculty and facilities needed to implement this project. However, to ensure consideration of minority organizations as defined in the Gray Amendment, we will work with the Office of Acquisition and Assistance Management, and the Office of Small and Disadvantaged Businesses, to identify all potential organizations and institutions which may be available to work with UH/CTAHR in the area of biological nitrogen fixation..

Anson R. Bertrand
Agency Director,
for Food and Agriculture
Bureau for Science and Technology

LFrederick:MEM:5/1/86:BNF4:Revised 5/30/86

WANG 3992g

SECTOR COUNCIL FOR AGRICULTURE

RECORD OF S&T PROJECT REVIEW COMMITTEE MEETING

COMMITTEE MEETING DATE: May 6, 1986

1. Project Office: S&T/AGR/RNR PID [ ] PP [xx] Project No: 936-4177

Project Title: Improved Biological Nitrogen Fixation thru Biotech

Proposed Contractor: University of Hawaii

Proposed Project Period: Sept. 30, 1986 - Sept. 30, 1996

Proposed Budget Period: same (000) Budget: $ 9.69 S&T/AGR 10,30MMissions/RBs $19.99

(If any, prior total est. cost: -0-)

(Grand total after adding this action: $19.99 Million

2. The members of this committee, and their findings are specified below:

Office Symbol	Name/Signature	Date	Endorsed	Not Endorsed
AFR/TR/ARD	Marc L. Winter	5-6-86	✓	
ASIA/TR/ARD	Charles Antholt	5/6/86	✓	Subject to priority sector issues handled at prior Sector Council discussion 5/6/86
BIFAD/s	John Stovall		✓	
LAC/DR/RD	Dwight Steen	5/7/86	✓	
ST/RD	Ken Swanberg	5/6/86	✓	
PPC/PDPR	Donald McClelland	5/6/86		
S&T/AGR	Tejpal Gill	5/6/86	✓	

3. It is the decision of this Committee that this project be:

✓ ENDORSED _____ NOT ENDORSED

SIGNATURE _____ Date _____

Anson R. Bertrand, S&T/FA
Chairperson

Any dissenting opinions are attached.

cc: S&T/PO, J. Holt

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AGRICULTURE SECTOR COUNCIL

Minutes of Meeting May 6, 1986

Attending:

Anson Bertrand, S&T/FA, Chairman
Dwight Steen, LAC/DR/Rd
Kenneth Prussner, AFR/TR/ARD
C.H. Antholt, ANE/TR/ARd
John Stovall, BIFAD/S
Donald McClelland, PPC/PDPR
Compton Chase-Lansdale, PRE/I
Tejpal S. Gill, S&T/AGR
Loren L. Schulze, S&T/AGR/AP
Kerri-Ann Jones, S&T/AGR/RNR
Mary Mozynski, S&T/AGR
Worth Fitzgerald, S&T/AGR/RNR
H.S. Plunkett, S&T/RD/RRD
Ken Swanberg, S&T/RD
Harvey Hortik, S&T/AGR/AP
C.H. Mullenax, S&T/AGR
Betty Roche, S&T/PO
Janine Finnell, S&T/EY
Edward Caplan, S&T/MGT, Executive Secretary

Outside Hires. In view of the Secretary of State's cable to embassies envisioning a reduced presence of the United States, the Sector Council decided to take no action at this time in regard to the selection of outside hires. Dr. Bertrand said agriculture is being left out in this area, as evidenced by Chuck Antholt's memo of April 9. The Secretary's cable, however, may result in AID cutbacks in the field.

Chuck Mullenax. Dr. Bertrand introduced Chuck Mullenax, an IPA and a veterinarian who has done a great deal of work in developing countries, mostly in Latin America. Dr. Mullenax briefed the Sector Council on his qualifications, saying his expertise is in forage-based livestock, the main problem of which is a decrease in the nutrient value of available forage. He offered his services to the regional bureaus.

Long-Term Training. The chairman reminded council members of Jay Morris's memo of April 21, which said a person may nominate himself or herself for long-term training. The council was also

reminded that Don Wadley has promised to detail for the council the procedure he used for assignment to long-term training.

American Soybean Association. Loren Schulze briefed the Sector Council on the status of the relationship between the Agency and the American Soybean Association, which has been critical of some of the Agency's activities in the past. Correspondence between the Administrator and the Association has resulted in a meeting between representatives of the two groups June 12-13. The Administrator sees the blossoming relationship as an opportunity to further his private sector initiatives and to turn a negative into a positive.

The meeting is to be informal, with a purpose of exploring items of mutual interest. AID has made up a tentative agenda for the afternoon of June 12, including an overview of the Agency, briefings on regional interventions regarding crops similar to soybeans, and discussions on the Agency's expectations of this meeting. The Association will soon determine an agenda for the morning of June 13. Schulze sees some sort of positive activity resulting from the meeting, an activity requiring cooperation of the Agency and the Association.

S&T Bureau CPSS. Dr. Bertrand reported that the newest draft of the CPSS was going to Dr. Brady today and that the drafting of Action Plans was going on simultaneously. He said it is possible that the Sector Council would need a meeting to discuss it.

Dr. Bertrand's Retirement. Dr. Bertrand announced that he is retiring effective June 30 and that Dr. Duane Ocker has been appointed Agency Director for Food and Agriculture, effective July 1. No one has yet been appointed director of the Office of Agriculture.

PID on S&T/AGR Irrigation Project. The Sector Council took up the PID on the Irrigation Management Support and Research project. Worth Fitzgerald explained that this project complies with the S&T Bureau's CPSS statement that there will be no new projects in S&T/AGR for 1987 because it is in essence a redesigning of the Water Management Synthesis II project. It will, in fact, carry the same project number and will be labeled a revision rather than a new project.

Fitzgerald provided a briefing on the 10-year project. The $40 million funding will come from AFR, LAC, S&T/AGR, S&T/RD and

S&T/EY. A working group has been revising the project since the need was first noted last July. The concept paper was approved by Dr. Brady and all bureaus have provided input.

A discussion ensued regarding a proposal by John Stovall that this project be designated a Title XII project. Several members disagreed with him, suggesting that private sector or other entities might fulfill at least part of the project requirements better than Title XII universities. It was determined that the decision over whether to designate the project for Title XII can be made at a later stage, and that was the council's consensus.

Stovall suggested that there are imaginative ways of working out combinations of private firms and universities to achieve the project's objectives. Betty Roche urged that time be taken to plan procurement and that the project not be limited by a contract as the sole means.

The PID was approved by all the bureaus except Africa, which abstained.

PP on S&T/AGR BFN Project. The Sector Council took up discussion of the project paper for the Improved Biological Nitrogen Fixation Through Biotechnology project. Tejpal Gill gave a briefing, terming the project's subject a technology that has come of age because of the high cost of nitrogen-based fertilizer. The $20 million project will be funded half from S&T/AGR and half from buy-ins. It will provide training, research, and technical assistance.

Compton Chase-Lansdale, saying the involvement of the private sector is critical to the success of the project, called attention to the lack of mention of investigation into commercial opportunities. He cited the possibility of biotechnology firms with proprietary interests, co-financing, and commercial opportunities in distribution. Several members expressed doubt as to whether private firms would want to get involved with the small quantities contemplated, but agreed that the idea should be investigated and that the results of the investigation be incorporated into the project paper. An advisory group from the private sector was also suggested.

The project paper was approved.

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Tab E

MAY 30 1986

MEMORANDUM

TO: M/SER/AAM, Ms. Kathryn Cunningham

FROM: S&T/FA, Anson R. Bertrand

SUBJECT: Non-Competitive Award of Cooperative Agreement
Improved BNF thru Biotech

I request that you consider only the College of Tropical Agriculture and Human Resources (CTAHR), Department of Agronomy and Soil Science (DASS), University of Hawaii (UH) for the subject cooperative agreement to: 1) utilize and enhance its resource base in international and tropical biological nitrogen fixation (BNF) developed over the past decade of cooperation with the A.I.D. and other donors; 2) expand the level and range of its collaboration with U.S., LDC, and regional public and private organizations and international institutions; and 3) increase its research activities in the area of BNF technologies.

The University of Hawaii is recognized world-wide for its organizational structure and its ability to implement a program of research, outreach and training in BNF. The organizational structure and qualifications of the University to implement BNF programs in tropical agriculture are listed below:

Organizational Structure

a. The University of Hawaii, by virtue of its location and unique environmental conditions, has a comparative advantage in the area of biotechnology research for tropical agriculture.

Tropical research is a very important element of the CTAHR and is included in various planning and development documents for the State of Hawaii. The CTAHR's unique capabilities in tropical agriculture were recognized by the Congress of the United States when it passed Section 406 of the Food for Peace Act in 1967 creating centers of Tropical Agriculture Research in the United States. Hawaii was one of the two original locations selected for this program. Currently more than 200 of the University's activities and programs have an international dimension.

The DASS in the CTAHR has had a strong international focus for more than sixteen years beginning with a series of strengthening grants (211d) from one of AID's predecessor agencies. These grants were established for the purpose of developing expertise in international tropical agriculture within the faculty of DASS. During the late 1960s and early 1970s, several areas of interest in the department were developed into exceptionally fruitful projects. These are International Benchmark Sites Network (IBSN) and Nitrogen Fixation by Tropical Agricultural Legumes (NifTAL). In addition, international and regional endeavors of the University of Hawaii include the following projects; Soil Management CRSP, Forestry/Fuelwood Resources Development, and South Pacific Regional Agriculture Development.

The DASS is a research department within the UH. It is administered by the CTAHR and conducts research in tropical soils and crops. CTAHR, in turn, is administered by the Office of the President of the UH through the Office of the Vice-President for Research and Graduate Education. CTAHR itself administers the Hawaiian Institute of Tropical Agriculture and Human Resources (HITAHR) which is responsible for overseeing the extension and research activities of all departments within CTAHR as well as running the State's experimental research stations.

b. NifTAL was established within the DASS at the University of Hawaii (UH).

For the past ten years, the NifTAL project has not only collaborated with other projects and members of the DASS in research and training activities related to nitrogen fixation technologies, but also interacted with the Departments of Horticulture and Microbiology in research and international technical assistance.

The NifTAL project has been a multi-disciplinary project since its inception. It draws on expertise from not only the DASS, but from all departments within the CTAHR largely through its research, training, and graduate student programs. It also collaborates with other projects within the DASS and with other departments in the CTAHR, the University of Hawaii Biotechnology Program, the Institute of Pacific Islands Forestry (USDA/FS), and with legume/BNF programs at CIAT, ICRISAT, IRRI, ICARDA, AND IITA.

Qualifications, including key personnel, facilities and equipment

a. The goal of the project is to: 1) increase crop productivity; 2) reduce inefficient use of imported agricultural inputs; and 3) encourage sustainable agricultural production systems. NifTAL has been accomplishing this goal through a three pronged approach to research, outreach including both technical assistance and networking activities, and a multi-tiered training program. Its staff, composed of highly skilled agronomists, microbiologists, soil scientists, and professional educators, conducts basic research to remove constraints to the fuller implementation of BNF technologies, coordinates research networks, and trains LDC scientists and policy makers in the various aspects of BNF technology.

b. Research Qualifications

The previous project, NifTAL, developed and maintains a rhizobium germplasm bank of over 1700 rhizobium strains for 283 legume species, including 150 fully characterized strains for the 50 most important tropical legumes. This comprehensive collection contains strains of rhizobia not only for crop plants with proven economic value, but also for legumes that are becoming increasingly more important in developmental programs, for example, tree and forage legumes. This resource is an invaluable source of germplasm for the studies planned in this project.

NifTAL also maintains a resource bank of antibodies to serologically identify the most efficient strains of rhizobia. The antibody collection covers 83 rhizobial strains which are used in legume production projects funded by the missions and LDCs. No other source of these antibodies presently exist.

Using these germplasm resources, NifTAL staff forms a strong research and development base to conduct BNF basic and applied research on: 1) selection, characterization, and evaluation of rhizobium germplasm for tropical legumes with actual or potential roles in development; 2) improvement of rhizobia using modern genetic technologies; 3) improvement of legume plant germplasm through mutagenesis and tissue culture; 4) development, production, and maintenance of a resource bank of antibodies required for the study of rhizobial ecology; 4) development of experimental designs and research networks to evaluate inoculation success; 5) development and testing of small and moderate scale inoculant production systems; 6) environmental determinants of inoculation success; 7) effects of management on nitrogen fixation and yields of tropical legumes; 8) maximizing nitrogen fixing potential of woody legumes; 9) selection and evaluation of rhizobia for the stresses of tropical environments; and 10) the economic benefits to the small LDC farmer of utilizing BNF technology.

c. Technology Transfer Qualifications

NifTAL has 10 years experience in technology transfer related to nitrogen fixation technologies. Its activities in this area include: 1) providing technical assistance to LDC scientists and institutions in the design of BNF research programs; 2) aiding government agencies in the assessment of overall legume programs and need for inoculant production capabilities; 3) helping in the design, and establishment of inoculant production plants; 4) developing and disseminating guidelines for inoculant quality control procedures and design of laboratories; 5) publishing newsletters, pertinent bibliographies. and specialized documents to assist LDC research and policy makers to become aware of, and stay abreast of, developments in BNF technology; and 6) evaluating ongoing LDC BNF programs, including quality of staff, equipment, facilities, and appropriateness of research goals.

d. Training Qualifications

The training program at UH/CTAHR is multi-tiered. Four graduate fellowships at the Ph.D. level are offered annually. Candidates from developing countries are accepted first by the University of Hawaii graduate school and once admitted conduct their doctoral research at the UH/CTAHR installation on Maui under the guidance of the NifTAL staff. The similarity of soils, climate, and crops grown in Hawaii and many LDCs make the University of Hawaii an ideal location for doctoral candidates from LDCs.

Short-term internships of from 3 to 6 months are also available. Candidates for this program, who are usually mid- or senior-level scientists, apply directly to NifTAL for admission to the program and work closely with the NifTAL staff during their stay.

In the past 10 years NifTAL has held nine technical training courses in basic BNF methodologies for BNF researchers, as well as courses in inoculant production, inoculant utilization, and management of nitrogen fixing trees for extension agents, policy makers and inoculant manufacturers. The majority of these courses were held in the developing countries or at the international agricultural research centers. The basic six-week BNF technology course has been conducted both at the NifTAL-Maui headquarters and in the LDCs. Because the specialized material required for this course is not in standard microbiology or agronomy courses, it was necessary for NifTAL to adapt standard materials for the lectures and laboratory research to the LDC conditions and environment to comply with the requirements of the participants. This adaptation will also be required under the new project and can only be accomplished by experienced BNF scientists with considerable knowledge of LDC agricultural production constraints.

e. Networking and Linkages

NifTAL developed and manages a network of over 200 scientists, extension workers, and policy makers interested generally in nitrogen fixation, and more specifically in inoculation technology. Most members, who come from 55 nations, were originally cooperators in the International Network of Legume Inoculation Trials Program (INLIT), whose primary purpose is to coordinate inoculation trials to determine if inoculation led to increased yields. NifTAL supplies the materials, experimental design, and statistical services required to run the trials. However, the network today not only facilitates research related to the INLIT trials, but also aids information transfer and communication between BNF researchers worldwide.

Initially the communication was largely two way between NifTAL and legume workers or LDC policy makers, with cooperators receiving NifTAL publications as well as INLIT materials. Later many of these individuals become involved in training programs and requested other services offered by NifTAL; e.g., rhizobium cultures, antibodies, research quality inoculants, and literature. Ultimately communication expanded to link individual network members both within and between countries. Several country networks of BNF workers now exist in many LDCs.

Administratively, the outreach section coordinates networking activities with NifTAL. One senior NifTAL staff member is designated a network coordinator for each of three regions; Africa, Asia and Near East, and Latin America and Caribbean. Each regional coordinator is responsible for identifying new collaborators as well as assuring that cooperators in his region receive: 1) the necessary materials for the inoculant trials; 2) all NifTAL publications; and 3) information on the other services and training that NifTAL offers.

The network coordinator is the key individual in the operation of the network. He is both the direct contact with the host country and USAID country mission personnel. Contact is initiated and maintained by personal visits, contacts at meetings and workshops, by correspondence, through a newsletter to trial cooperators, through regional meetings of cooperators, and through the BNF bulletin.

In addition to the NifTAL network of BNF researchers, extension agents, and policy makers in developing nations, NifTAL maintains active research and communication linkages with all major BNF laboratories in the United States and other developed nations and with the BNF/legume programs at the international agricultural research centers.

To identify the proper source to implement this project, S&T/AGR considered the following universities, which were rejected for the reasons cited:

a. North Carolina State University (NCSU) has extensive experience in international programs and nitrogen fixation research. While NCSUhas developed and delivered training courses overseas, NifTAL's multi-tiered approach to training is more suited to the needs of LDCs. NifTAL's courses are designed not only for LDC scientists, but also for administrators and policy makers. In addition, NifTAL offers the only hands-on BNF technology and inoculant production courses.

   NCSU has a large germplasm collection for a few crop species. However, this collection does not contain well characterized, genetically diverse rhizobia for tropical crop, tree and pasture legumes. N.C. State also lacks the complementary antisera collection required for the proposed environmental constraints program that is maintained at UH. Lastly, the geographical location of NCSU does not allow testing of new bacterial and plant germplasm under the climate and soil conditions that prevail in the tropics.

b. The University of Florida (UF) has several researchers active in nitrogen fixation work and a substantial international program in farming systems. However, missing is the appropriate environmental conditions necessary for the studies proposed, as well as overseas experience in BNF training courses and technology transfer. Finally, the lack of bacterial germplasm and tropical soils and climate would not allow successful completion of the proposed project.

c. The University of California at Davis has conducted significant research in the area of selection of rhizobia for the stresses found in many tropical environments. However, this institution lacks experience in international development, technology transfer, and conducting training courses overseas. Finally, the lack of tropical environmental conditions would make it impossible to conduct some of the proposed research at Davis.

The most compelling reason for the selection of the University of Hawaii over other institutions is the fact that it has a major and concerted effort entirely devoted to BNF by tropical legumes. Although each of the above three institutions has several faculty members working on BNF, the momentum and the critical mass of scientists that has been assembled at the University of Hawaii are unique to tropical agriculture.

Recommendation:

It is for the above reasons that the Office of Agriculture, the Directorate for Food and Agriculture, and the Bureau for Science and Technology recommend that the Office of Acquisition and Assistance Management award a Cooperative Agreement to the Department of Agronomy and Soil Science in the College of Tropical Agriculture and Human Resources, University of Hawaii.

Clearances:	S&T/AGR,	Tejpal Gill	_____	date _____
		Frances Li	(draft)	date 5/16/86
		Lloyd Frederick	_____	date _____
	S&T/PO,	Betty Roche	_____	date _____
		Gerald Gower	_____	date _____

BNF5:WANG:4115g:LFrederick:MM:5/20/86

Tab F

MAY 30 1986

MEMORANDUM

TO: M/SER/AAM, Ms. Kathryn Cunningham

FROM: S&T/FA, Anson R. Bertrand

SUBJECT: Award of Companion Basic Ordering Agreement
Improved BNF thru Biotech

I request that you negotiate only with the University of Hawaii's College of Tropical Agriculture and Human Resources (CTAHR), Department of Agronomy and Soil Science (DASS) for a companion basic ordering agreement to the cooperative agreement also being processed at this time for the subject project. This request is based on Section 6.302-3 of the Federal Acquisition Regulations (FAR) for the exemption of a non-competitive agreement which cites: full and open competition need not be provided for when it is necessary to award the agreement to a particular source or sources in order to establish or maintain an essential engineering, research, or development capability to be provided by an educational or other nonprofit institution or a federally funded research development center.

Identification of the Agency and agreement activities

The Department of Agronomy and Soil Science (DASS) is a section of the College of Tropical Agriculture and Human Resources (CTAHR) with long term and well established collaboration (staff sharing, joint curricula and research projects) in teaching, research and service work - including international research and development, technical assistance, training and networking.

The cooperative agreement will fund basic and adaptive research at the University of Hawaii, provide for training opportunities, networking activities, technology transfer, and backstopping mission funded projects. The Special Orders under the Basic Ordering agreement to be funded by missions, regional bureaus and AID/W offices will provide for the following:

Short- and medium-term technical and advisory services will:

- Identify constraints imposed on BNF by the stresses of tropical soils.
- Monitor microorganism/plant interaction related to BNF in the LDCs.
- Field test BNF technologies in the LDCs that have been developed at the University of Hawaii.
- Conduct workshops and training programs based on the BNF technologies developed at the University of Hawaii.
- Plan, organize, reorganize and implement BNF inoculant production and supply programs or projects and their integration into the overall LDC strategy for agricultural development, including extension services, contract farmers and parastatal and private organizations.

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- Identify, analyze and recommend solutions to managerial, financial and operational programs in rhizobium inoculant production.

- Monitor agriculturally important nitrogen-fixing microorganisms introduced into the soil environment as inoculant for economically important legumes.

- Gather environmental data of important crop and tree legumes inoculated with rhizobium.

- Assist in the establishment of BNF regional resource centers in the LDCs.

Long-term assistance will:

Provide assistance and the scientific data required for the successfully operation of the regional resource centers, including:

- Long-term testing of research results developed at the UH.

- Conducting short-term and non-degree biotechnology training courses and co-sponsor workshops on specific topics of research.

- Assisting scientists to conduct applied research experiments which were designed at UH.

- Assisting in mission BNF projects using techniques developed at UH.

- Promoting and facilitating the establishment of small BNF businesses using the techniques developed at UH.

- Publishing regional newsletters and information bulletins to disseminate research data developed at UH and the regional centers to extension agents and country researchers.

The experience gained from the activities funded under special orders against the companion basic ordering agreement will be fed directly back into the activities funded under the cooperative agreement; i.e., UH's research agenda, training curricula, research network linkages, and technical transfer and informational services. It is also intended that the occasion for Mission funded activities shall arise from work financed under the cooperative agreement. Mission funded tasks may be identified by the cooperator and approved by missions and the Office of Agriculture in the course of the cooperator's engagement in institutional strengthening activities. Alternatively, UH may receive requests for assistance directly from the Office of Agriculture, regional bureaus, missions and LDC public and private organizations.

The relationship between the cooperative agreement and the Basic Ordering Agreement is outlined in the attached project paper, Section 5.0, Method of Implementation on pages 25 - 27.

Recommendation:

It is for the above reasons and in accordance with section 6.302-3 of the FAR that the Office of Agriculture, the Directorate for Food and Agriculture and the Bureau for Science and Technology recommend that the Basic Ordering Agreement be awarded to the College of Tropical Agriculture and Human Resources of the University of Hawaii.

Clearance:	S&T/AGR:Tejpal Gill	_____	Date	_____
	Frances Li	(draft)	Date	5/16/86
	Lloyd Frederick	_____	Date	_____
	S&T/PO,Betty Roche	_____	Date	_____
	Gerald Gower	_____	Date	_____

BNF6:WANG 4115g:S&T/AGR:LFrederick:MEM

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

Revised 4/29/86: Revised 5/8/86

Improved Biological Nitrogen Fixation thru Biotechnology

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Revised 4/30/86:5/8/86

Improved Biological Nitrogen Fixation Through Biotechnology

Acronym Listing

AID	Agency for International Development
BNF	Biological Nitrogen Fixation
BOSTID	Board on Science and Technology in Development
CIAT	International Center for Tropical Agriculture
CRSP	Collabotative Research Support Program
CSRS	Cooperative State Research Service
FAO	Food and Agriculture Organization, UN
H_2	Hydrogen
IARC	International Agriculture Research Centers
IBPGR	International Board for Plant Genetic Resources
ICRISAT	International Crop Research Institute for Semi-Arid Tropics
IEE	Initial Environmental Examination
INF	Infection
INLIT	International Network of Legume Inoculation Trials
IRRI	International Rice Research Institute
LDCs	Less Developed Countries
MIRCEN	Microbiological Resources Center
N	Nitrogen
N_2	Nitrogen Gas
NH_3	Ammonia
NIF	NifTAL Strain
NifTAL	Nitrogen Fixation by Tropical Agricultural Legumes
NC	North Carolina State University
NSF	National Science Foundation
P	NifTAL Rhizobium Strain
PID	Project Identification Document
PP	Project Paper
PSTC	Program in Science and Technology Cooperation (SCI)
SCI	Office of the Science Advisor
S&T/AGR	Office of Agriculture, Bureau for Science and Technology
UH	University of Hawaii
UNDP	United Nations Development Programme
USDA	United States Department of Agriculture
USDA/ARS	United States Department of Agriculture, Agricultural Research Service

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Improved Biological Nitrogen Fixation (BNF) Through Biotechology

Preface

The Office of Agriculture, Directorate for Food and Agriculture, Bureau for Science and Technology recommends that $19,990,000 be authorized for a new ten-year project, "Improved BNF Thru Biotech". It is also recommended that this project be implemented under a Cooperative Agreement and Basic Ordering Agreement with the College of Tropical Agriculture and Human Resources, Department of Agronomy and Soil Science, University of Hawaii (UH). Of the total $19,990,000, $9,690,000 will be provided by S&T/AGR for the core budget and $10,300,000 is expected to be provided by missions, regional bureaus and AID/W offices to support Regional Resource Centers, participant training, and research and other activities directly related to the research and development activities to be funded under the core budget. In addition, the UH is expected to contribute another $2,468,000 in services and equipment to achieve the purpose and goal of this new project.

The PID for this project was approved by the Senior Assistant Administrator for Science and Technology on March 20, 1986. It has been identified by S&T/AGR as being of high priority, and the funding is included in the S&T/AGR budget plans. The plan of action presented in this recommendation will provide substantial benefits to LDC countries to increase their food production with minimum inputs. The cost estimates are appropriate for the planned level of effort.

The project will promote the use of biological means to fix nitrogen in the soil increasing nutrients for plants. The availability of more nutrients increases the quantity and quality of food available to the LDCs. The project workplan is a multi-pronged design that involves research, training, information, networking, technical assistance and private enterprise. Products that emerge through research and development (Program 1 - Genetic Technologies and Program 2 - Methods for Monitoring Microorganisms) will be field tested under (Program 3 - Environmental Data Collection) involving the Regional Resource Centers (Program 4). Use of this technology by LDC farmers will be facilitated through an incentive system by way of small LDC commercial plants receiving technical assistance by NifTAL under Program 5.

Nitrogen is an essential constituent of proteins, and large amounts are required by all forms of life. Other forms of life derive their protein from plants. For non-legume crops, synthesis of their protein depends on nitrogen compounds present in the soil. The amount and availability of the soil nitrogen to plants is conditioned by biological processes in the soil.

Nitrogen and other essential nutrients removed from the soil must be replenished, in one way or the other, to ensure sustained agricultural productivity. This can be accomplished through the addition of chemical fertilizers which are expensive and require heavy capital investment for production, storage, transportation and application. For example, the industrial synthesis of nitrogen requires large quantities of natural gas from fossil fuel plus additional energy to maintain the high temperatures and pressures necessary to chemically combine nitrogen gas (N_2) with hydrogen (H_2) to form ammonia (NH_3). Although ammonia can be used directly, it must be processed further to produce a solid fertilizer suitable for use by small LDC farmers. This additional step requires additional energy and increases the cost.

In contrast to this, there are symbiotic microorganisms that can satisfy the nitrogen needs of plants by directly incorporating into their proteins nitrogen gas (N_2) which is the major constituent of air. This process, in which plants overcome the stress of nitrogen deficiency through the use of microorganisms to provide the necessary nitrogen),is referred to as biological nitrogen fixation (BNF) While this process occurs naturally, it can be greatly improved by a number of methods.

The first approach, and one which NifTAL has concentrated on to date, is the identification of well suited rhizobia for a given legume under specific conditions. This approach is termed selection. Using this method NifTAL has acquired a comprehensive collection of rhizobia specifically adapted to tropical conditions. These strains are characterized and available for distribution to researchers throughout the world. This research, in addition, to training, field testing, technology transfer, and networking provides the foundation for the new proposed project which is needed to further improve and maximize the use of BNF in the developing world.

The new project will build on the established foundation to advance our understanding and control of BNF. It will do this by using traditional methods, as well as the new methods of biotechnology. These new methods allow the problems to be approached on a molecular level. Through genetic engineering nitrogenase genes or symbiotic genes can be manipulated leading to the development of improved microorganisms. Additionally, more sophicated methods for monitoring the bacteria/plant interaction will be developed and the environmental conditions which affect this interaction will be more accurately defined. This project also plans to adapt and transfer the technology by providing support to regional centers and commercial inoculation production. For additional scientific and technical information, please refer to Appendix D - State of the Art, and Appendix E - Technical Review.

1.0 Project Rationale, Perceived Problem and Background

1.1. Project Rationale

Nitrogen is the most common limiting nutrient needed to increase agricultural production in the tropics. This nutrient can be obtained from commercial fertilizers or through biological means. The high cost of fertilizer makes it

virtually impossible for small LDC farmers to obtain and use on their fields. An alternative is for the farmers to adopt agricultural practices that will increase nitrogen concentration in the soil through biological means. Biological nitrogen fixation (BNF) is an economical way to supply substantial amounts of nitrogen to the soil to increase food production. This new project will give special attention to the development and utilization of biotechnologies related to nitrogen fixation applicable to tropical soils and agriculture with potential returns to temperate agriculture.

1.2 Perceived Problem

The world is currently operating at about 40 percent capacity in terms of potentially arable land for food production. The largest portion of this land, 1.6 billion hectares, is located in the tropics and includes about 0.8 billion hectares of land considered marginal for agricultural use. Major strategies for solving the world food problem must involve not only increasing yields in already cultivated lands, but also increasing the world's cultivated areas.

Food production technology is highly sophisticated in the developed countries of the temperate region. This production technology is not necessarily suited to the climate, soil and economics of the tropical environments of the developing countries. The high input, high technology farming methods employed on large farms in developed nations rarely meet the needs of LDC farmers who cultivate less than one or two hectares of land and do not have the scientific knowledge to use sophisticated methods. To break the hold of poverty, the LDC farmers must produce more food more efficiently to increase real per capita income.

The quality of life of the rural people in many developing countries is bound to the quality and productivity of the land. Most of the less-developed countries are located in the tropics. The challenge is to provide more food with higher nutritional quality and a better life for an ever-increasing population on a fixed land area characterized by impoverished soils.

1.3 Background

In FY 1974, an unsolicited proposal for the predecessor project, N-Fixation, Symbiotic, was received from the University of Hawaii to develop and disseminate a technology to optimize the use of BNF in tropical agricultural legumes. A contract was signed in FY 1975 with the College of Tropical Agriculture and Human Resources, Department of Agronomy and Soil Science, University of Hawaii (UH) to establish an interdisciplinary unit called Nitrogen Fixation by Tropical Agricultural Legumes (NifTAL), dedicated to the application of technologies based on biological nitrogen fixation (BNF) to contribute to the international development goals of the Agency.

NifTAL's early years were spent building a resource capability in Hawaii to service the needs of national programs of the developing countries for research support, information and technical services, and multi-tier training.

In 1979, NifTAL initiated global, regional, and in-country networks to enable developing country scientists to draw on the substantial body of knowledge already available to answer key research questions. These networks have been effective in providing developing countries with access, not only to the resources represented in NifTAL, but also to expertise of leading BNF scientists throughout the U.S., Europe, and Australia.

Initially, NifTAL dealt only with tropical grain legumes important in human nutrition. Later, the tropical forage and tree legumes were included. This expansion is consistent with NifTAL's role as a flexible and responsive unit, offering development support in a key process (BNF) rather than a specific discipline or commodity. This approach balanced and complemented the legume programs of the international agricultural research centers (IARCs), Collaborative Research Support Programs (CRSPs), and others, which are committed to specific commodities and/or defined geographic areas.

The BNF program is coordinated through the efforts of the S&T/AGR project officer utilizing the lead scientists (Principal Investigator, NifTAL) of the University of Hawaii. The lead scientist of the UH project participates in all coordinating meetings, and has excellent rapport with the USDA scientists working with the Rhizobium Germ Plasm Bank in Beltsville, Maryland, and the lead scientst (principal investigators) of the research grants under the Biotechnology Limiting Factors small grants project under agreement with the Cooperative State Research Service (CSRS). Under this small grants project, U.S. and developing scientists work collaboratively to overcome impediments to the effective use of biological nitrogen fixation.

The principal investigator of the UH project helps find appropriate visiting scientists for training at Beltsville, and incorporates the results of research in the Biotechnology Limiting Factors project into the suggested BNF package of practices. The UH scientists are involved in consulting with the recipients of grant under the Office of the Science Advisor (SCI) program, and have organized workshops for the BNF network of scientists. The book "Biological Nitrogen Fixation Technology for Tropical Agriculture" published in 1982 by CIAT, was the result of one of these workshops. These coordinated efforts enable researchers in the U.S. and the LDCs to search for ways to overcome factors important in developing countries that limit the use of biological nitrogen fixation.

Originally, NifTAL aimed its training at key researchers through postgraduate programs, internships, and short courses. Later training efforts were expanded to extension leaders and LDC manufacturers of commercial inocula.

In February 1985 the predecessor project was reviewed by a NSF team of scientists who recommended continuation of the activities with UH to: 1) maintain a rhizobium germplasm resource and a serum bank housing antisera to identify the strains of rhizobium used in inoculants for the International Network of Legume Inoculation Trials (INLIT); 2) determine the yield benefit resulting from inoculation of local varieties; 3) develop systems for improving the inoculant delivery; 4) develop improved techniques and cultural

practices; 5) acquire multiple stocks of seeds of target legumes; 6) study the effects of soil infertility; 7) provide economic information on the relative costs of the use of BNF technology; 8) provide regional and in-country support; and 9) train professional researchers, technicians, and extension workers. A copy of the review teams report is available upon request from the Office of Agriculture. For additional information, please refer to Appendix E, Technical Review.

The rhizobia germplasm collection is a major resource for LDCs. It includes more than 1700 strains from 285 different legume species which were collected from 56 countries. Three superior strains for each of 16 major tropical legume crops were characterized, antisera prepared against each strain and research grade rhizobia inoculant provided to LDC researchers on request. More than 200 scientists in the third world have participated in coordinated inoculation trials to increase crop yields.

Trained scientists and technicians in legume-based BNF technologies are in short supply in the third world. Training is a major component of this new project. LDC scientists and technicians will be trained at UH, regional resource centers, national centers and IARCS. Approximately five short courses will be offered annually for an estimated 103 LDC participants. Intern trainees at the University come from all areas of the developing world where soil fertility is a major problem. Approximately eight graduate students are scheduled annually for advanced degrees at the University. Technical assistance includes site visits, personal correspondence, publications and bulletins. Under the previous project NifTAL serviced approximately 750 requests annually for documents, including 378 reprints of NifTAL work.

A further evolution in NifTAL was the regionalization of development support. In 1983, NifTAL established a BNF Resource Center in Bangkok, Thailand to provide research support, technical and material services, and training oriented specifically at crop production constraints of the South and Southeast Asian region. The Director of the Center is supplied by NifTAL. Through he efforts a national workshop was held for extension leaders on the use of legume inoculants in Bangladesh. This workshop was funded by the Bangladesh Agricultural Research Council. Other countries in the region are planning similar workshops to be implemented by the University of Hawaii under the new project.

2.0 Project Goal, Purpose and Description

2.1 Project Goal

The goal of this project is to: 1) increase crop productivity; 2) reduce inefficient use of imported agricultural inputs; and 3) encourage sustainable agricultural production systems.

2.2 Project Purpose

The purpose of this project is to: 1) increase the efficiency of nitrogen fixing microorganisms adapted to LDC conditions through methods of biotechnology; 2) promote the use of BNF in LDCs by assisting them to adapt, use, and disseminate information about BNF; and 3) increase their capacity to produce and distribute BNF inocula.

2.3 Project Description

Increases in crop productivity have been considerable from the use of BNF as demonstrated by the NifTAL project, IARCs and individual scientists in the U.S. and developing countries. In a sense, the successes so far have opened up new challenges. The following major areas still require research before the full potential of BNF in farmers fields can be realized: i.e., 1) scientific problems in establishing effective nitrogen-fixing symbioses; and 2) mechanisms for effective transfer of proven biotechnologies to LDCs.

Scientific problems with BNF technologies can be further sub-divided into the following categories: 1) lack of success of conventional research methods to establish effective nitrogen-fixing symbioses on traditional LDC legume crops; and 2) lack of knowledge about appropriate BNF technologies for new legume crops, principally trees.

To date, soybeans introduced into new areas have exhibited consistent, positive responses to inoculation. Dramatic yield increases have been the result. Unfortunately, the inoculation response of basic food staple legumes, such as dry beans in Latin America, cowpeas in Africa, and chick peas and other pulses in Asia has been highly variable.

One of the most important reasons for lack of inoculation response is the inability of inoculum strains to effectively compete with indigenous rhizobia for nodulation sites on host plants. The traditional approach to overcome this problem has been to select more competitive inoculum strains. This is largely a trial and error process that is very site specific and has little predictive utility. Consequently, LDC farmers of traditional crop legumes cannot know whether or not inoculation will be beneficial.

A new approach is needed for implementing tested BNF biotechnologies and transfering these technologies to the farmers. To date, little attention has been focussed on stimulating the private sector in LDCs to promote BNF technologies. In several instances, proven BNF technologies exists which could benefit small farmers if made available to them. Lack of promotion prevents these technologies from being utilized. The experience of NifTAL and others involved in biotechnology transfer has been that LDC organizations designed to promote agrotechnologies, if existent, are often underfunded or ineffective. In terms of both the technology and the level of local interest, the time is ripe to enlist the energies of the LDC private sector in the production and use of materials and techniques for BNF.

Recently identified legume crops, such as trees, are increasing in importance in development programs. As a group, their biology is poorly understood. Even less is known about their requirements for microsymbionts. However, it is safe to assume that because of the limited fertilizer resources available to the LDC farmer, tree legumes will have to depend heavily on microsymbionts, such as rhizobia, mycorrhizal fungi, and Frankia for essential nutrients.

This project encompasses research, networking, training, technology transfer and regionalization of biotechnology resources. Research will be the major emphasis totalling 45 percent of the ten-year budget and will involve use of modern genetic methods and modeling techniques to solve scientific problems. The service component will total 55 percent and will include training, technology transfer and networking. Assistance will be provided through the regional outreach program to promote small business incentive programs.

This project will consist of five separate programs which will be supported by S&T/AGR, University of Hawaii, missions, LDCs, regional bureaus and other AID/W offices.

Program 1	Genetic technologies for Improvement of Rhizobium/Legume Symbiosis for Crops and Trees;
Program 2	Development of Methodologies for monitoring Microorganisms Introduced into the Environment; and
Program 3	Environmental Data Collection to Maximize Performance of Biological Nitrogen Fixation;
Program 4	Regional Resource Centers; and
Program 5	Commercial Inoculant Technical Assistance

The five programs together form an integrated approach for optimizing the impact of BNF technologies. The "better" rhizobia produced under Program I will be field tested in Program 3 using the methods developed in Program 2. This testing will be supervised in part by the appropriate Regional Resource Centers (Program 4). Implementation of the technology (production and use) will be facilitated by small LDC enterprises assisted under Program 5. In addition, the "better" rhizobia will be made available to other scientists for evaluation under the Biotechnology Limiting Factors small grants program.

2.3.1 Research Components

The research activities will be designed to maximize the amount of nitrogen gained from symbiotic nitrogen fixing systems. Problems imposed by the inherent genetic potential of the bacteria and the host plant will be addressed using modern genetic engineering tools as well as more conventional biotechnology methods. Constraints imposed by the stresses of tropical oils will be identified through global research networks and LDC scientists. The data base generated by the network will be used to develop a predictive model to inoculate rhizobia used for important crop and tree legume production.

The proposed research activities between U.S. and LDC scientists will emphasize using modern genetic methodologies to engineer rhizobia and host plants for tropical environments (Program 1). One crop to be highlighted in this research is nitrogen-fixing trees. Other crops to be selected will be those important to the participating countries, regional centers and IARCs. The research efforts with this crop will produce optimized tree/microbe symbioses that can be integrated into development projects for forage, fuelwood and other tree products.

Complementary to obtaining improved microorganisms (Program 1) is evaluating their persistence (Program 2) and performance (Program 3) in the field, to permit the development of a predictive inoculation response model.

2.3.2 Networking Components

Networking is a major part of this project. To accomplish the objectives in Programs 3 and 5 requires a functional network to develop a predictive model of inoculation response using data from only one or a few sites. This activity will be closely coordinated with the Biotechnology Limiting Factors small grants program under agreement with USDA/CSRS to attain a greater degree of interaction and collaboration than is normally found between isolated grantees. Similarly, the formation of a small business network may facilitate interaction and more efficient problem solving. A network narrative and listing of cooperators are attached as Appendix G.

2.3.3 Training Component

All programs are designed with training components, including degree, technical training and workshops and seminars.

Degree Training	Program 1	Genetic Technologies
	Program 3	Environmental Data Base
Short courses & workshops	Program 3	Environmental Data Collection
	Program 1	Genetic Technologies
	Program 2	Monitoring Methods
	Program 5	Commercial Inoculant Technical Assistance

In addition, senior LDC scientists will spend time at the project headquarters or at the Regional Resource Centers conducting collaborative research with appropriate UH staff. These Centers will provide opportunities for degree and technical short-term training. Non-degree training programs in BNF technologies will be offered to key personnel responsible for providing technical backstopping to production-oriented legume/rhizobium symbiosis programs. NifTAL's connection to small scale farmers and collaborators is attached as Appendix C.

The research results of LDC scientists working collaboratively with NifTAL have been published in scientific journals. A partial listing is attached as Appendix K. In addition, in late 1985, a questionnaire was sent to LDC participants of NifTAL's training courses and others receiving the BNF Bulletin Newsletter. The responses received to date are summarized in Appendix L.

2.3.4 Technology Transfer (Outreach Program)

All programs are designed to deliver to the LDCs validated technologies developed at the University of Hawaii. The genetic technologies for improvement of rhizobium/legume symbiosis for crops and trees (Program 1), the methodologies for monitoring microorganisms introduced into the environment (Program 2), and the environmental data identified under Program 3 will be transfered to the LDCs through participant training programs, short courses, workshops, and dissemination of research findings through BNF bulletins, literature reviews, etc. The BNF Resource Centers will provide additional mechanisms for transferring research results and methodologies developed under the various programs to extension leaders and to the LDC farmers' fields. In addition, technical services will be provided to missions, LDCs and IARCs upon request and as funded by the requesting institution or office. This activity will establish personal and professional relationships essential for sustaining research progress and technology transfer.

2.3.5 Regionalization of Biotechnology Resources

Many LDCs have some degree of BNF expertise and technology, but practically all of them need additional technical support and encouragement. The greatest potential for increasing NifTAL's effectiveness in reaching its goals would appear to rest in establishing Regional Resource Centers in strategic points within LDC countries where there is concrete evidence of local scientific, political, and financial support. A center has already been established in Thailand by the Thai Ministry of Agriculture. This center provides a strong foundation for research, technology transfer, technical assistance and an innovative and cost-effective method to train professional and technical agriculturalists in scientific methods appropriate to local needs. Other centers are being planned which will allow BNF technologies to be adapted to an array of soil, legume, and climatic variables while strengthening local institutional capabilities. The Thailand Center was visited by the NSF evaluation team, who reported that it appears to be an excellent model for other Centers to use. The backstopping for these regional centers will be funded under the core budget and will provide the needed support to the Centers. The funding for the Centers, however, must come from the missions or regional bureaus.

The Africa Bureau is planning a Regional Center in Dakar, Senegal for genetic upgrading of nitrogen fixing tree species. The University of Costa Rica has submitted a request to the University of Hawaii to establish a BNF Resource center on the campus. Another center is being planned for Morocco. These centers will act as liaisons between NifTAL, Hawaii and LDC individuals and institutions. They will: 1) provide a strong foundation for research, training, technology transfer, and technical assistance; 2) publish quarterly newsletters to disseminate to the LDC farmers scientific information on nitrogen fixing microorganisms; 3) assist regional and national seed projects to increase crop production for food, oil, and agroforestry products; 4) design and implement production and distribution systems; 5) assist in establishment and operation of small businesses for production and/or marketing of inoculum; 6) provide to scientists selected rhizobial cultures and inoculants which are adapted to regional soil constraints; 7) provide technical back-up to Agricultural Development Officers in the development and implementation of in-country programs; and 8) work closely with the Biotechnology - Limiting Factors Small Grants Program.

2.3.6 Commercial Inoculant Production Technical Assistance

This program is designed to reach the small LDC farmers with the latest BNF technology through an incentive system that benefits not only the user, but also the producer and distributor of inoculant. A multi-sector developmental and promotional approach will be implemented. After successful trials have identified the rhizobia inoculant for a specific environment, UH will contact U.S. private sector investors to outline the potential benefits of inoculant production in that environment.

The developmental phase will include: 1) research to modify the technology to make it more adaptable for tropical conditions and appropriate for LDC applications; and 2) training to produce a cadre of personnel capable of applying the technology. The promotional approach will include information dissemination and successful demonstration of the benefits of the new BNF technologies. A successful model has been developed in Turkey where sufficient capital was raised through private investments and loans to construct a inoculant production plant. For additional information on this program, please refer to Appendix E, pages 5, 6 and 7.

3.0 Project Programs, Outputs, and Inputs

This project, Improved BNF thru Biotech, will assist the University of Hawaii to expand its activities in the following programs:

3.1 Project Programs

The following programs will be funded under this new project:

3.1.1 Genetic technologies for improvement of rhizobium/legume symbiosis for crops and trees

Rationale for funding: A number of characteristics of the bacteria/plant interaction determine whether optimum biological nitrogen fixation occurs. These include the following: 1) the formation of the nodule; 2) the effectiveness of the nitrogenase enzyme system; 3) the ability of the bacteria to compete and survive in the soil among other bacteria; and 4) the ability of the plant and the bacteria to manage the harsh soil conditions (e.g., salinity). These characteristics are governed by specific plant or bacteria genes and it is the purpose of this program to identify and manipulate these genes in order to produce the best possible BNF system. A germplasm collection is extremely important in this endeavor since a resource of beneficial traits is a necessity.

The objective of this program is to maximize the amount of nitrogen fixed by the symbiotic system using modern genetic tools, as well as more conventional biotechnological approaches. It will use the genetic characteristics for stress tolerance that are already present in tropical rhizobia to engineer better strains for tropical legumes. Research activities will include:

1) Genetic improvement of rhizobium for tropical legumes, including:

   a) Introduction of host-range and symbiotic plasmids for specific tropical legumes into rhizobia from NifTAL's extensive germplasm and other collections that have already been identified as superior in competitiveness and soil-stress tolerance (e.g., TAL 1145, USDA 123, CIAT 899, NC 92, and P320) to develop superior strains for other tropical legumes.

   b) Development of strains that harbor multiple copies of "Nif" structural genes, and Infection (Inf) genes, through collaborative endeavors with scientists in Mexico, Thailand, and Pakistan.

   c) Evaluation of the benefit of multiple Nif- and Inf-genes in controlled hydroponic systems, to be developed, that would maximize plant growth so that its symbiotic potential is taxed and realized.

   d) Identification and isolation of genes involved in competition from rhizobia already recognized as supercompetitors (e.g., TAL 1145, USDA 123) and transfer of these genes into other rhizobia for tropical legumes.

   e) Utilization of the genetic background of adapted indigenous strains from stressful soils, to produce more effective and competitive rhizobia.

   f) Genetically construct multi-pupose rhizobia by introducing into them cloned genes for other beneficial traits, i.e., B.T. insecticides from Bacillus Thuringensis or root protectants from pseudomonas.

2) Biotechnological approaches for improving host performance of the following:

   a) Crop legumes, to include:

      - Evaluating plant materials produced by the "Tissue Culture for Crops" Project with Colorado State University for yield performance and symbiotic potential under tropical soil/climate conditions; and

      - Mutagenesis breeding to develop tropical crop legume lines analogous to those successfully developed for soybeans and peas. Screening and selecting mutant lines for:

        - Supernodulation;
        - Nitrate insensitivity;
        - Erectoid growth habit (e.g., in winged beans); and
        - Non-nodulation for experimental assessment of BNF by legumes.

   b) Tree legumes, to include:

      - Collaborating with scientists in the Philippines and Costa Rica on tissue-culture selection of tree/microsymbiont symbioses suited to the stresses of tropical soils.

3) Germplasm resources, to include:

   a) Maintaining NifTAL rhizobium culture collection;

   b) Assembling symbiotic plasmids and cloned genes for use by NifTAL scientists and LDC collaborators.

   c) Selecting and testing strains for adverse and stressful environments;

   d) Evaluating materials produced in the genetic research program;

   e) Maintaining seed collections of tropical legumes of potential importance in development;

   f) Propagating and maintaining seeds of legume species, particularly trees, that are neglected or endangered;

   g) Distributing rhizobium and seeds to LDC and IARC scientists for research and inoculum production; and

   h) Preparing teaching modules on NifTAL's rhizobium technology, and courses on inoculum production.

4) Training developing country scientists.

3.1.2 Development of methodologies for monitoring microorganisms introduced into the environment

The ability to monitor the fate of bacteria added to the soil is critical. Improvements in soil survival or ability to nodulate are indicators in the BNF process which must be monitored in order to determine progress.

The objective of this program is to develop and evaluate methods for monitoring agriculturally important nitrogen-fixing microorganisms introduced into the soil environment as inoculant for economically important legumes.

Activities under this component will include:

1) Producing antisera and maintaining an antiserum bank;

2) Distributing and producing antisera upon request by LDC and IARC scientists;

3) Developing antibodies for characterizing important tropical serogroups;

4) Developing genetic markers for rhizobial strains for which serological methods of identification are inadequate;

5) Training LDC scientists in monitoring methodologies; and

6) Producing teaching modules on monitoring methodologies.

3.1.3 Environmental data collection to maximize performance of biological nitrogen fixation

The objective of this program is to gather an environmental data base and use it to develop a predictive model for response of important crop and tree legumes to inoculation with rhizobium.

The activities under this program are:

1) Selecting a set of sites from the INLIT Program and IBSNAT/Benchmark Soil Project;

2) Identifying former NifTAL trainees and other qualified developing country scientists from NifTAL's INLIT program to carry out standardized experiments;

3) Holding a workshop to design a uniform series of experiments which will generate the minimum data set needed to build the inoculation response model;

4) Characterizing experimental sites physico-chemically and microbiologically;

5) Contracting with collaborators to run the experiments and collect the minimum data set;

6) Holding regional workshops on the protocol for the standardized experiments;

7) Conducting experiments to correlate the persistance of rhizobia introduced into amended and control plots with site characteristics;

8) Performing studies to identify major abiotic constraints to plant growth and nitrogen fixation;

9) Conducting uniform inoculation field trials to generate information on the interactions between management, inoculation, and environment;

10) Encouraging and assisting in publication of data generated by collaborators;

11) Using the data collected from activities 4), 7), 8), and 9) construct a predictive model; and

12) Validating the model in sites where the minimum data set was collected, as well as in new sites.

3.1.4 Regional resource centers

The objective of this program is to maintain and strengthen the BNF Regional Resource Center in Bangkok, Thailand and establish at least two other regional centers; one in Africa (e.g., Kenya or Senegal) and one in Central America (e.g., Honduras or Costa Rica) using the highly successful Thailand center as a model. Additional information is contained in Appendix D, pages 19, 20 and 21.

The activities of these centers could include the following:

1) Perform adaptive research of results produced under programs 3.1.1, 3.1.2 and 3.1.3.

2) Conduct Short-term and non-degree biotechnology training courses and co-sponsor workshops on specific topics related to programs 3.1.1, 3.1.2, 3.1.3 and 3.1.5;

3) Assist scientists (visiting the Centers for periods of up to 12 months) to conduct collaborative research with Center personnel;

4) Contact USAID and other donors in the region to coordinate activities, assist in mission project design, and identify opportunities to incorporate BNF technologies into USAID projects;

5) Collaborate with IARCs to extend BNF technologies to crops and regions not reached by the international centers.

6) Promote and facilitate the establishment of small biotechnology businesses in the region by acting as an intermediary between developing country entrepreneurs and NifTAL, Hawaii;

7) Monitor activities of Program 3.1.3 being implemented in the region;

8) Promote NifTAL services and training courses; and

9) Publish regional newsletters and information bulletins for dissemination to extension agents and country researchers.

3.1.5 Commercial Inoculant Technical Assistance

This program will provide training and technical information, with the objective of encouraging developing country entrepreneurs U.S. private investors to establish small biotechnology businesses in the LDCs. The activities listed below will be funded primarily from the delivery orders issued under the Basic Ordering Agreement with UH.

The activities will include:

1) Providing technical backstopping to support small LDC entrepreneurs and private U.S. firms.

2) Training to produce a cadre of personnel who are capable of applying the biotechnologies and will promote and market these biotechnologies.

3) Contacting scientific and business leaders in developing countries to assess interest/potential for particular product such as rhizobium inoculants (while doing other activities under the programs listed in 3.1.1., 3.1.2., and 3.1.3);

4) Conducting regional training workshops on the technical aspects of "How to set-up a rhizobium inoculant business;"

5) Conducting regional technical courses on "Inoculum Production Biotechnology;"

6) Producing promotional flyers on specific products and their uses;

7) Providing superior strains and materials for production of high quality inoculants;

8) Establishing procedures for quality control testing and a voluntary incentive program for securing a quality "Seal of Approval;" and

9) Publishing a manual for developing country entrepreneurs which contains information for at least 10 countries on "How to Establish Small Rhizobium Inoculant Businesses in Developing Countries."

10) Identifying potential sites for U.S. private investment and contacting appropriate U.S. private sector investors for expression of interest and participation.

11) Contacting the Bureau for Private Enterprise to facilitate start-up procedures and funding for inoculant production.

3.2 Project Outputs

The following outputs are anticipated to be accomplished by the end of the project. The time frame when these outputs are to be achieved is shown in the Critical Performance Indicator table attached as Appendix B.

3.2.1. Genetic technologies for improvement of rhizobium/legume symbiosis for crops and trees

1) Genetically improved rhizobium strains

a) Superior rhizobia tailored for important tropical legumes and matched with specific environments;

b) Competitive rhizobia strains able to overcome the barrier presented by indigenous rhizobia;

c) Rhizobia improved in nitrogen fixation and nodule formation due to presence of multiple Nif and Inf Genes;

d) Rhizobia better adapted to prevailing stresses of tropical soils; and

e) LDC graduate students and post doctoral scientists trained in modern genetic concepts and methodologies.

2) Improved host legumes

a) Characterized plant genotypes that grow better and fix nitrogen in stressed environments;

b) Beneficial mutants of tropical legumes with increased nodulation and enhanced nitrogen fixation even in the presence of high levels of soil N;

c) Erectoid mutants of crops with potential promise in the tropics; i.e., winged bean;

d) Non-nodulating mutants of important tropical tree legumes essential as reference plants in studies assessing BNF potential of leguminous trees;

e) Tree/rhizobia symboises tolerant of tropical soil and climatic stresses; and

f) LDC graduate students and postdoctoral scientists trained in agricultural biotechnology.

3) Germplasm

a) A comprehensive collection of rhizobia for tropical legumes maintained;

b) Germplasm made available to LDC scientists, inoculum producers and other worldwide collections;

c) Three field-tested strains for 15 of the most widely used legumes (from INLIT Program) made available for research and application;

d) Stocks of high quality, peat-based rhizobium inoculants for priority legumes available for distribution. Inoculants for other legumes prepared on request;

e) Catalogue of all strains held in NifTAL's Rhizobium Germplasm Resource continue to be made available;

f) Catalogue of NifTAL's Legume Germplasm Resource prepared and distributed; and

g) Seeds of legumes recommended for specific soils and agroclimatic conditions distributed to LDC collaborators for testing and use.

3.2.2 Development of methodologies for monitoring microorganisms introduced into the environment

1) A resource bank of antibodies for microorganisms of agricultural importance.

2) Resource germplasm of microorganisms characterized for genetic markers and intrinsic antibiotic resistance.

3) Several research publications in refereed journals and abstracts in conference proceedings.

4) Five visiting scientists trained at NifTAL.

5) Two biotechnology training courses held annually, each of six weeks duration.

6) Published training manual on monitoring methodologies. Distribution will be primarily to LDC researchers.

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7) Collaborative linkages on biotechnology established with LDC institutions.

8) Antisera and genetically marked organisms provided to LDC scientists.

3.2.3 Environmental data collection to maximize performance of biological nitrogen fixation

1) A model that can predict inoculation response by tropical legumes as a function of environment and management practices.

2) Regional networks composed of developing country scientists that can engage in other scientific activities of regional importance.

3) Linkages among regional networks to form a global network of LDC scientists with shared problems and common goals.

4) Enhanced capacity of LDC institutions to solve important agricultural problems involving BNF uses.

5) Developing country professionals trained at M.S. and Ph.D. levels.

6) Twenty publications disseminated.

3.2.4 Regional Resource Centers

1) Bangkok Center strengthened. New centers will be established in Morocco, and in one country in Africa and Latin America.

2) A quarterly Regional Resource Center Newsletter will be distributed from each of the three Centers.

3) LDC scientists using NifTAL strains and materials and providing feedback.

4) BNF technologies incorporated into mission crop production projects.

5) Training courses planned in collaboration with Programs 3.1.1, 3.1.2, and 3.1.3. Whenever possible courses will be jointly-sponsored with other donors.

6) One to two visiting scientists per year conducting collaborative research at each Center. Costs paid by visiting scientist home institution whenever possible.

7) Assistance provided to at least two small biotech companies in each region with Center Assistance.

8) Several scientific articles published in refereed journals based on work by visiting scientists.

3.2.5 Commercial Inoculant Technical Assistance

1) Inoculant production plants established in several LDCs

2) Small private enterprises incorporated and operational

3) High quality inoculum with seal-of-approval available for marketing and sale to individual consumers, in-country programs, and government seed producers.

4) Publications describing technical set-up of production plants and logistics of starting and doing business in specific LDCs.

5) Motivated, trained entrepreneurs operating commercially viable biotechnology ventures.

3.3 Project Inputs

The following inputs will be required to implement this project:

3.3.1 Genetic technologies for improvement of rhizobium/legume symbiosis for crops and trees

1) Facilities, equipment and supplies for genetic manipulation and testing of rhizobia.

2) Staff
- One senior level scientist with expertise in genetic methodologies and/or plant sciences
- One mid-level (M.S.) technical scientists
- Two graduate assistants, from LDC, when possible
- One postdoctoral fellow, from LDC when possible
- One technical assistant
- Administrative support

3.3.2 Development of methodologies for monitoring component activities

1) Facilities, equipment and supplies
- Laboratory, greenhouse, field and animal facilities
- Laboratory equipment; i.e., ultra-centrifuge
- Miscellaneous laboratory supplies

2) Staff
- One senior level microbiologist
- One research associate
- Administrative support

3.3.3 Environmental data collection to maximize performance of biological nitrogen fixation

1) Equipment, supplies, freight costs,

2) Staff
- Senior network coordinator
- Biometrician (0.25 FTE)
- One research associate
- Two graduate student assistantships
- Administrative support

3) Other Costs
- Publications
- Travel
- Networking

3.3.4 Regional Resource Centers

1) Facilities and Equipment
- Rental of office and laboratory space at hosting institution
- Operational funds

2) Staff
- Senior Regional Resource Center Coordinator at UH
- For each Center the following:
  - Senior scientist outposted in each regional center (Center Director)
  - Associate Center Director (M.S. or Ph. D.)
  - Research Assistant (B.S. or M.S.)
  - Clerical
  - Accountant/Information specialists

3) Other Cost
- Travel

3.3.5 Commercial production technical assistance

1) Back-up technical services and assistance from NifTAL, Hawaii

2) Staff
a) University of Hawaii Small Business Coordinator

b) Each Regional Resource Center
- Regional Resource Director (.50 FTE)
- One Senior Program Coordinator
- Local consultants
- One information/publications specialist
- Administrative support

4.0 Cost Estimate, Financial Plan and Person Months

4.1 Cost Estimates

The total cost of this ten-year biotechnology subactivity is estimated at $19.99 million, which includes an S&T/AGR core budget of $9.69 million, and an expected $10.30 million contribution from the missions, regional bureaus and other AID/W offices under the Basic Ordering Agreement. The participating LDC institutions must cover the local costs of the Regional BNF Resource Centers. In addition, the University of Hawaii is expected to contribute another $2.468 million to achieve the goal and purpose of this project. These cost estimates are based on the calculated level of effort needed; i.e., facilities, personnel, travel and allowances, supplies and services and other requirements necessary to achieve targeted outputs of the types and magnitude as set forth in the logical framework (Appendix A).

The annual line item budgets are included in the Logframe, Appendix A. The summary budgets are listed below:

4.1.1 Summary Obligational Schedule - A financial summary table is shown below:

Table 1
(In thousands)

	FY 1986	FY 1987	FY 1988	FY 1989	FY 1990	FY 1991	Total Five-Yr	Total Ten-Yr.
S&T/AGR	$ 200	$ 960	$ 960	$ 990**	$ 960	$ 760	$ 4,830	$ 9,690***
Buy-Ins *		1,050	1,125	1,400	1,350	1,350	6,275	10,300
Total AID	$ 200	$2,010	$2,085	$2,390	$2,310	$2,110	$11,105	$19,990
U of Hawaii	-	305	338	206	214	171	1,234	2,468
Grand Total	$ 200	$2,315	$2,423	$2,596	$2,524	$2,281	$12,339	$22,458

* Includes funding from missions, regional bureaus and other AID/W offices from a combination of programmed and unprogrammed sources and will be implemented by Delivery Orders under the Basic Ordering Agreement issued in conjunction with the Cooperative Agreement funded by S&T/AGR.

** Includes $30 thousand for an indepth evaluation.

*** Includes $90 thousand for three indepth evaluations.

4.1.2 Summary Budget Tables - The following tables summarize the total five-year and ten-year budgets by Project Component (Table 2a and 2b) and by Line Item Expenses (Table 3a and 3b). For the five-year budget, the funding of the research component from all sources is estimated at 53 percent; technology transfer at 20 percent; training, including degree and non-degree training, workshops and short-term courses at 22 percent; and networking at 5 percent.

The University's contribution in cost-sharing over the initial five year cooperative agreement is estimated at $1.234 million or just a little under 26 percent of S&T/AGR's contribution.

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Table 2a
Five-Year Summary by Project Components
(in thousands)

Components	Cooperative Agreement S&T/AGR	(%)	Hawaii	Total	Delivery Orders Buy-Ins	Total	Percent
Research	$2,160	(45%)	$ 687	$2,847	$3,765	$ 6,612	53%
Technology Transfer	960	(19%)	195	1,155	1,255	2.410	20%
Training	1,200	(25%)	234	1,434	1,255	2,689	22%
Networking	480	(10%)	118	598	-	598	5%
Evaluation	30	1%	-	30		30	
Total	$4,830	(100)	$1,234	$6,064	$6,275*	$12,339	100%

* Under the Delivery Orders of the Basic Ordering Agreement, the University of Hawaii shall perform only the research and those technical assistance services which apply the methodologies, systems and policy findings developed under the Cooperative Agreement.

Table 2b

Ten-Year Summary by Project Components

(in thousands)

Components	Cooperative Agreement S&T/AGR	(%)	Hawaii	Total	Delivery Orders Buy-Ins	Total	Percent
Research	$4,320	(45%)	$1,374	$5,694	$6,210	$11,904	52%
Technology Transfer	1,920	(19%)	390	2,310	2,070	4,380	19%
Training	2,400	(25%)	468	2,868	2,020	4,888	26%
Networking	960	(10%)	236	1,196	-	1,196	2%
Evaluation	90	1%	-	90	-	90	-
Total	$9,690	(100)	$2,468	$12,158	$10,300*	$22,458	100%

* Under the Delivery Orders of the Basic Ordering Agreement, the University of Hawaii shall perform only the research and those technical assistance services which apply the methodologies, systems and policy findings developed under the Cooperative Agreement.

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Table 3a

Five-Year Summary costs by Line Item Expenses
(In thousands)

Components	Cooperative Agreement S&T/AGR	Hawaii	Total	Delivery Orders Buy-Ins	Total
Salaries/Wages	$2,525	$ 500	$ 3,025	$ 2,400	$ 5,425
Benefits	505	105	610	759	1,369
Consultants	-	-	-	1,290	1,290
Travel	195	-	195	376	376
Publications	220	-	220	81	301
Supplies	280	-	280	376	656
Total	$3,725	$ 605	$4,330	$5,282	$ 9,612
Indirect Costs(20%)*	745		685	835	1,580
Equipment	95	629	724	-	724
Training	60	-	60	158	218
Utilities	175	-	175	-0-	175
Total CA & BOA	$4,800	$1,234	$6,034	$6,275	$12,309
Evaluation	30	-	30	-	30
Grand Total	$4,830	$1,234	$6,064	$6,275**	$12,339

Table 3b

Ten-Year Summary costs by Line Item Expenses
(In thousands)

Components	Cooperative Agreement S&T/AGR	Hawaii	Total	Delivery Orders Buy-Ins	Total
Salaries/Wages	$5,050	$1,000	$ 6.050	$ 3,450	$ 9,500
Benefits	L,010	210	1,220	1,081	2.301
Consultants	-	-	-	2,620	2,620
Travel	390	-	390	650	1,040
Publications	440	-	440	119	559
Supplies	560	-	560	751	1,311
Total	$7,450	$1,210	$8,660	$8,671	$17,331
Indirect Costs(20%)*	1,490		1,490	1,403	2,893
Equipment	190	1,258	1,448	-	1,449
Training	120	-	120	226	346
Utilities	350	-	350	-0-	350
Total CA & BOA	$9,600	$2,468	$12,068	$10,300	$22,368
Evaluation	90	-	90	-	90
Grand Total	$9,690	$1,468	$12,158	$10,300	$22,458

* Indirect costs of staff, travel, publications and supplies needed for the Regional Centers are computed at 14%.

** The funding levels for the delivery orders are based on projected requirements for assistance directly related to the research and development activities to be funded under the cooperative agreement with UH.

4.2 Annual Projections by Person-Months

The following table lists by activity, discipline and/or type of employment the person-months to be provided by S&T/AGR and UH under this project. Note it is estimated that S&T/AGR will fund a total of 210 person-months per year; whereas Hawaii plans to contribute another 24 person-months from its own resources for the same period to assure successful achievement of the goal and purpose defined in the Logical Framework, Appendix A. The University of Hawaii personnel (scientists and others) and the qualifications of the key personnel charged to the project are listed in Appendix H. Scientists and experts which are available for consultation, but not charged to the project are listed in Appendix I. In addition, a listing of the laboratory, equipment, and other facilities to be used by the project, but not charged to the project is included in Appendix I.

Table 4

Person Months per Year

	S&T/AGR	U Hawaii
Staff Support		
Director, NifTAL		12
Senior Scientists		12
Agricultural Economist	3	
Secretary	12	
Clerical	12	
Information/Publication Specialists	12	
Field Workers (3)	36	
Total Staff Support	75	
Program 1 - Genetic Engineering		
Genetic Methodologist/Plant Science Expert	12	
Technical Person Mid-Level (M.S.)	12	
Graduate Assistant (2)	24	
Postdoctoral Fellow	12	
Total Program 1	60	
Program 2 - Methodologies for Monitoring Microorganisms		
Microbiologist	12	
Research Associate	12	
Total Program 2	24	
Program 3 - Environmental Constraints		
Senior Network Coordinator	12	
Research Associate	12	
Biometrician	3	
Total Program 3	27	
Program 4 - Regional Resource Centers		
Senior Regional Resource Centers Coordinator	12	
Program 5 - Small Business Promotion Entrepreneurship		
Senior Small Business BNF Coordinator	12	
Total Annual Person-Months	210	24

5.0 Method of Implementation - This project will be implemented under a five-year Cooperative Agreement with the University of Hawaii and will cover the cost of the core staff funded by S&T/AGR and the "Buy-ins" by Special Orders under a Basic Ordering Agreement. These Special Orders will be funded by the missions, regional bureaus and other AID/W offices. The predecessor, centrally funded project on Biological Nitrogen Fixation has an excellent success record. The long-term funding commitment of the University of Hawaii is therefore considered assured with the approval of this new project.

Through S&T/AGR and the University of Hawaii funding of the activities listed in 3.0 above, a critical mass of multidisciplinary scientists (see Table 4 above) will be available to backstop the Biological Nitrogen Fixation requirements of the missions. In addition, the core funding will provide the necessary administrative support to implement the project components.

Listed below are missions expected to participate in this project. This listing is based on information obtained from individual mission cables, data available at the University of Hawaii, completed questionnaires which were completed by missions, and discussions with the regional bureaus.

Table 5

Possible Missions to Participate in this Project

Regional Bureau	USAID	Remarks
Africa Bureau	Botswana	Mission has indicated a need for training in nitrogen fixing trees in agroforestry
	Burundi	Possible interest, beans are a major crop
	Cameroon	Possible interest, legumes are a major crop
	Kenya	Possible interest, beans are a major crop
	Mali	Possible interest, cowpeas are a major crop
	Mauritania	Mission has indicated a possible need for training activities
	Malawi	Bean & Cowpea CRSP has a program
	Niger	Possible interest. cowpeas are a major crop
	Rwanda	Mission indicated possible need for technical assistance. In addition, beans are a major crop.
	Senegal	A Regional Resource Center is being planned for Senegal. BNF methodologies are being used to improve soil fertility
	Tansania	Bean & Cowpea CRSP has a program
	Zambia	Mission indicated need for training, technical assistance in rhizobia inoculant production, and a $150,000 sub-contract with University of Hawaii.
	Zimbabwe	Mission indicated interest in N-Fixation to improve soils. Beans are a major crop.

Table 5

Possible Missions to Participate in this Subproject

Regional Bureau	USAIDs	Remarks
Asia and Near East	Bangladesh	To increase BNF in the farming systems program by providing inoculum strains, short-term consultants, training courses, and assistance on fermentors, azolla research, agroforestry and legume trees.
	Burma	Technical Assistance is needed on inoculant production
	Egypt	Agr. Research Center has given priority to BNF and coordinates the BNF research in the country.
	India	BNF research involves some of the foremost Indian and U.S. scientists, and strong linkages have been built between the 2 countries.
	Indonesia	International legume inoculum trials are being performed in Indonesia.
	Jordan	The ADO officer, Bill Furtick, has expressed interest in BNF
	Morocco	Hassan II Institute of Agronomic and Veterinary Sciences is very interested in establishing and operating a "Regional Resource Center" for nitrogen fixation biotechnology.
	Nepal	The ADO officer has indicated BNF work is important and training will be required.
	Pakistan	Follow-up on the assessment of the BNF research.
	Philippines	BNF research is being conducted at the University of Philippines at Los Banos
	Sri Lanka	Limited BNF activities are carried on in the country.
	South Pacific	The mission indicated the need for symbiotic nitrogen fixation technologies in tropical legumes and cycling of nitrogen in stable cropping systems.
	Thailand	A BNF Regional Resource Center has been established in Bangkok to facilitate biotechnology training, promote small legume inoculum production businesses and disseminate information on the use of biological methods to fix nitrogen in the soils.
	Tunisia	A Biotechnology - Limiting Factors Grant is being implemented in Tunisia by Oregon State in collaboration with Tunisian Scientists

Table 5

Possible Missions to Participate in this Project

Regional Bureau	USAIDS	Remarks
Latin America and Caribbean	Belize	Mission has indicated a possible need for BNF training course for extension workers, and a willingness to collaborate in BNF activities.
	Bolivia	Mission indicated possible need for training, absolute need for technical assistance and a willingness to collaborate in the new BNF initiatives.
	Costa Rica	Dr. Trejos (University of Costa Rica) has indicated an interest in establishing a regional center.
	Dom. Republic*	Mission indicated possible need for training, and technical assistance, and a willingness to collaborate with the new initiatives in BNF.
	Guatemala*	Mission indicated a possible need for training in nitrogen fixing trees and technical assistance.
	Haiti	Mission indicated a positive need for all types of training, and technical assistance, and a willingness to collaborate with the new initiatives in BNF.
	Honduras*	Dr. Malo, Director, Zamorano has indicated to UH an interest in establishing a Regional Resource Center
	Jamaica	Mission indicated possible need for training 1-2 people at UH, willingness to collaborate with the new initiatives in BNF and use an IQC and cost sharing for activities in BNF.
	Peru	Nitrogen fixation activities are currently conducted by Carlos Arbizu of the Universidad San Christobal de Huamanga, Ayacucho. The main interest is in tropical N-Fixing trees, small grain legumes and forage legumes (temperate and tropical).

* Countries where the Bean and Cowpea CRSP is operating. In addition, the Bean and Cowpea CRSP has programs in Brazil, INCAP and Mexico.

Note: Food crops which are grown in Latin America and the Caribbean include phaseolus vulgaris (dried field bean), cowpeas, pigeon peans and peanuts. Tree legumes include leucaena, profopis and liricidia. Alfalfa is grown in the upland range country. NifTAL is currently working on these food, tree and range crops and this work will continue under the new project.

6.0 Implementation Plan

Upon project approval, a brief worldwide information cable will be sent to the missions informing them of the implementation plan and requesting information on the needs of the missions during FY 1987. Thereafter, similar cables will be sent to missions in August of each project year to obtain information for the University of Hawaii for its annual work plan.

6.1. S&T/AGR Project Management Responsibility

This project will be managed by the S&T/AGR project manager who will deal directly with the University of Hawaii to provide technical and administrative support and oversight. Current AID staff and procedures are adequate to implement this project. The project manager will spend approximately 60 percent of the time managing the activities to assure that the objectives are achieved, facilitate acquisition of mission requests and funding, monitor project operations, approve certain administrative actions within the project, maintain close liaison with the grantee, and communicate regularly with the Principal Investigator, University of Hawaii. Specifically, the project manager also will be responsible for the following actions:

- Prepare for the UH a listing of mission projects dealing with nitrogen-poor soils or depleted soils. This list will be updated annually in time to permit UH to develop the annual work plan to meet the needs of the missions, regional bureaus and international centers.
- Prepare for UH a listing of all S&T projects which are related to biological nitrogen fixation or which generate data that may be used by UH in the implementation of research and development activities.
- Maintain contact with regional bureaus and missions to identify and expedite receipt and response to requests for assistance in biological nitrogen fixation.
- Communicate directly with the Principal Investigator at UH on matters related to mission requests or other information required in the performance of the cooperative agreement and the delivery orders under the Basic Ordering Agreement.
- Establish liaison with other AID project managers in project areas set forth in Section 3.0 above and assist UH to develop relationships with the project contractors, grantees and participating agencies.
- Approve scientists and consultants hired by UH under the Cooperative Agreement.
- Monitor project operations by assuring that the biological nitrogen fixation reports meet the reporting requirements as set forth in Section 7.2 below.
- Schedule and coordinate project evaluations and management reviews.
- Clear travel requests for UH scientists and experts traveling to LDCs.
- Approve the annual work plan, including research to be undertaken.
- Monitor progress of the annual work plan and overall program giving special attention to keeping the project on track.
- Maintain contact with the external panel of BNF scientists and supply the members with the data required to provide advise on matters concerning research, administration, training, technical assistance and networking.

6.2 UH Management Responsibility and Qualifications

The scope of work of this project is entirely in the area of biological nitrogen fixation where the UH has particular and unique implementation expertise. It does not require the assignment of new functions or activities nor the expansion of an existing function or activity which would increase capital investment or annual operating costs at UH. It is administratively part of the Department of Agronomy and Soil Science of the College of Tropical Agriculture and Human Resources. The Department Chairman and the Dean of the College take a strong interest in the project and actively assist the Principal Investigator when called upon.

The BNF activity is integrated with College-wide projects in an actively coordinated multidisciplinary program. Tenure track positions will be created for the scientists to enhance the integration of the program with the University, the College, and the Departments of Agronomy and Soil Science. University equipment and research facilities will be used to the extent possible as part of the cost-sharing under the Cooperative Agreement. The project will draw on the diverse resources of the other departments of the University of Hawaii. UH scientists and facilities which are available for the project, but not charged to the project are listed in Appendix I.

6.2.1 UH Management Responsibility

The Principal Investigator (PI) has the overall responsibility for implementing the project. The PI draws on advice from two senior advisors of the College of Agriculture on policy relating to agronomic or microbiological aspects of the project's scope of work. The PI is supported by an Associate Project Director with primarily responsibility for administration of the project's headquarters, operations in Hawaii and its overseas activities. Specifically the PI will have responsibility for implementing the following functions:

- Design the annual work plans in collaboration with the S&T/AGR project manager and obtain approval prior to implementation. The annual work plans will outline the activities to be implemented under Section 3.0 above.
- Maintain close working relationships and communicate regularly with the S&T/AGR project manager, regional bureaus, missions and IARCs.
- Develop and implement the required reporting structure to conform with the project reporting requirements.
- Oversee the timely completion of all reports set forth in the reporting requirements described in Section 7.2 of this project paper.
- Obtain clearance from the S&T/AGR project manager to hire scientists and consultants to implement the activities in this project paper.
- Obtain approval of the requesting missions prior to sending scientists and other professionals to the mission.
- Carry out the annual work plans within the time frame and financial resources allocated.
- Coordinate the work of the UH headquarters with the work of the regional centers.
- Approve activities proposed by the regional centers

6.2.2 UH Qualifications

The proposed ten-year project is based on the successful BNF research conducted by Hawaii since June 1975. The new cooperative agreement will assist the University to expand its resource base and collaborate with U.S., LDC and international institutions in biological nitrogen fixation. The areas to be covered under the Cooperative Agreement are: 1) using genetic technologies to rhizobium/legume symbiosis for crops and trees; 2) monitoring microorganisms, 3) gathering data to identify environmental constraints; 5) assisting in establishing and/or maintaining regional resource centers; and 5) helping LDC entrepreneurs establish rhizobia inoculum production and distribution businesses in the private sector.

The State of Hawaii has always had a special interest in the development of tropical agriculture due to its geographic location in the tropics. The University, in turn, has been heavily involved in tropical agricultural research for over half a century. In particular, the University has worked actively in tropical legumes and rhizobial microbiology, the key scientific subjects of this project, for over thirty years prior to 1975 when the first contract (AID/ta/C-1207) on Symbiotic Nitrogen Fixation by Tropical Legumes was negotiated with Hawaii. The combination of geographic location and long-term prior interest in the subject research were vital factors in the selection of Hawaii for the initial three-year contract. Research in biological nitrogen fixation by tropical legumes requires field testing and field experiments in a tropical environment because of the sensitivity of the nitrogen fixing organisms to high soil temperature, to the tropical wet/dry climate cycles and to soil acidity found in tropical soils. The combination of all these factors is found only in a tropical environment. The only other major U.S. research institution in the tropics is the University of Puerto Rico. However, it, in contrast to Hawaii, has neither the depth, nor breadth of experience in the subject area of research, nor sufficient qualified staff to conduct the subject research. The southern tip of Florida has a subtropical environment with occasional winter frosts; the soils there are nonacidic and therefore, not typical of the acid soils of most tropical LDCs. Florida, therefore, is less suitable for the subject research.

In addition to the on-site tropical environment for field testing, the University of Hawaii has substantial investment in facilities and specialized equipment required for this project. In addition to Hawaii's laboratories, field testing plots, and scientific equipment, all suited for tropical agriculture research and procured with state funds over the years, other improvements and acquisitions have been made with AID funds and are specially tailored for this project. A greenhouse has been built for testing and laboratories set up and specially equipped as research and training facilities. An empty school, provided by the County of Maui, Hawaii for this project, has been renovated to meet specific project requirements. These facilities are vital for this research effort and would have to be duplicated at AID's expense if another contractor was chosen for the new cooperative agreement.

The International Network of Legume Inoculation Trials (INLIT) Network of over 200 LDC scientists will form the pool from which collaborators for networking activities will be selected. These individuals are ideally suited to participate in project programs because of their previous work with rhizobia and familiarity with the basic methodology required and interest in project goals. Hawaii has assembled a staff of experts from interrelated disciplines, who were carefully selected on the basis of their scientific ability and previous tropical experience. The primary and basic focus of this Hawaiian staff has been, and continues to be, on tropical agriculture whereas other U.S. institutions have only limited exposure to tropical agriculture. The following key senior Hawaiian scientists working on this project are considered predominant experts in their respective areas of specialization .

Scientists	Area of Specialization
Dr. B. Bohlool, Director	Tropical rhizobium ecology, bacterial genetics
Dr. Joann Roskoski	Tropical plant ecology, plant physiology
Dr. Paul Singleton	Agronomy, plant physiology

6.3 External panel of BNF scientists and a representative from PRE

An external panel of BNF scientists and a representative from PRE will be established to advise the S&T/AGR project manager on matters concerning administration, research, technical assistance, training, networking and private sector involvement of this project. The panel members will consist of representatives from the following:

- Biotechnology - Limiting Factors Project
- Biotechnology - Tissue Culture project
- One of the regional BNF centers; i.e., Thailand Regional Center
- Either ICRISAT or CIAT
- Bureau for Private Enterprise

The activities of the Panel will be supported by the project. Specifically, the Panel will assist in the following areas:

1) Review the annual work plans and recommend additions, modifications, or deletion of components, as appropriate.

2) Consult with the UH Principal Investigator on the management implications of the suggested modifications.

3) Participate in the annual reviews and the indepth evaluations of the activities under the Cooperative Agreement.

4) Identify opportunities for U.S. and LDC private sector involvement in inoculant production and distribution.

6.4 Country Selection Criteria - The country selection criteria for participation in this project will be based on the following:

1) Capacity of host country scientists to participate in the research and extension work; and

2) The degree of mission and regional bureau support.

3) Likelihood of socioeconomic acceptance of recommended practices;

4) The suitability of poor and depleted soils by upgrading by biological methods;

5) Prior identification of suitable sites by expert groups on the regional importance of the use of BNF methodologies;

6) Probability of high benefit/cost ratios for increasing crop production;

7.0 Monitoring Plans - Monitoring will be accomplished by the Principal Investigator, the S&T/AGR Project Officer, the Agricultural/Rural Development Officers of the participating missions; and the cooperating host government agencies. Annual progress reports and copies of all documents resulting from field and laboratory activities will be submitted to the Project Officer for clearance prior to distribution.

7.1 Annual Work Plans - UH will develop the annual work plan as a working document to guide the operations and achievements expected from the project. It will be forwarded to S&T/AGR for approval each year. The first plan will be submitted to S&T/AGR no later than 30 days after the cooperative agreement is signed and will cover the period October 1, 1986 to December 31, 1987. Therefore, the annual work will be due 60 days prior to the end of the calendar year; i.e., November 1.

S&T/AGR will review the contents of the proposed work plan with the external panel of BNF scientists, ask for points of clarification, if required, and grant final approval of the contents as proposed or modified by agreement between UH and S&T/AGR. This process of review and approval will be completed not later than 30 days after receipt of the original work plan from UH.

The annual work plan shall include the following:

- A list of activities to be undertaken (including private sector initiatives) during the year, categorized by project components; i.e., research, technology transfer, training and networking.
- A statement of how the activities relate to the output and research priorities.
- A projected beginning time frame for execution of the activity.
- A projected ending time frame for completion of the activity.
- The projected expenditure of person-months of input for each activity.
- The projected stage of activities at the end of the work plan or the projected outputs at the end of the work plan.
- Specific qualifications which may be required for certain activities given that many of the activities within the project are predicated on the amount of mission funding to be obligated for project activities.
- Notation of the specific activities planned to emphasize assistance to Africa.
- Baseline data on pricing, policy, marketing and agricultural inputs to the extent necessary to update the economic analysis to determine the yield benefit resulting from inoculation of local trials.
- Attachments to the work plan may include, but not be limited to the following: critical performance indicators, specific activity reports, and time qualification conditions.
- Methods to collect economic data on the relative costs of using BNF technologies.

7.2 Reporting Requirements - In addition to the Annual Work Plans described above, UH will submit the following reports within the specified time frame. These reports will provide pertinent data for S&T/AGR to monitor project activities.

7.2.1 Technical and Research Reports - Technical research activities of the project will be summarized in reports and distributed to the appropriate missions, LDCs and IARCs to encourage use of the technology developed. Normally such reports will be completed 60 days after completion of the activity. Journal articles and other external publications are encouraged. Manuscripts should be submitted to the S&T/AGR Project Manager prior to submission to a publisher as well as a copy of the resulting publication.

7.2.2 Annual Activity Reports - An Annual Report of the UH's international biological nitrogen fixation activities will be prepared. Although principally a technical document, it nevertheless must include pertinent statistics on quantitative information regarding the project and its activities described in Section 3.0 above. An Impact Analysis Report (as defined in Section 7.2.5 below) will be appended to this report which will be considered an instrument for technology transfer. A minimum of five copies should be submitted to the S&T/AGR Project Manager within 90 days of the end of each project year.

7.2.3 Training Activities - Summary of training activities undertaken under and in conjunction with this project is required annually, including the numbers of trainees by gender, nationality, training site, type of training activities, duration and purpose.

7.2.4 Annual Expenditure Reports - UH will submit annual expenditure reports by: 1) project line item; and 2) estimated distribution by project component, i.e., research, training, technology transfer and networking. The format will be collaboratively developed by S&T/AGR project manager and the Principal Investigator at UH.

7.2.5 Impact Analysis Report - An annual report will be submitted as an annex to the annual activity report (Section 7.2.2 above) which summarizes the impact of UH activities in the public and private sector in terms of increased crop production from the use of biological nitrogen fixation methods. This will provide a feed-back system for measurement and evaluation of the impact of services and training provided.

The impact analysis is defined as a measurement of results generated by activities undertaken by UH in accordance with the project description in the subject project paper and the scope of work in the Cooperative Agreement. For the most part, the impact analysis will be qualitative in nature, and quantified only as appropriate and will cover activities funded under this project and/or the previous contracts with UH.

7.2.6 Trip Reports - Trip reports will be prepared for each TDY assignment or trip to an LDC. The report will contain, but not be limited to, the following information: 1) logistical information, i.e., type of activity, geographical area of activity, dates of TDY, and team composition; 2) objective of TDY, including scope of work, as appropriate; 3) activities performed while on TDY; 4) summary of any technical reports resulting from TDY; 5) summary of identifiable techniques or information which could be transferred to other LDCs; and 6) summary of future potential needs of, or opportunities for, assistance to LDCs or missions, including possible networking potential. One copy of this report will be forwarded to S&T/AGR not later than 30 days after the staff member returns to UH. The trip report generally will not exceed 4 pages.

7.3 Reviews and Evaluations

7.3.1 Management Reviews

Management reviews will be conducted annually by the S&T/AGR project manager in consultation with UH, missions, regional bureaus and other institutions, as appropriate. Since such reviews may involve visits to UH or specific LDCs, the review process will be funded by S&T/AGR. Such annual reviews will be undertaken after submission of the annual reports as described in Section 7.2 above. The annual reports (activity and impact) will become an integral part of the review process.

7.3.2 In-depth Evaluations Three formal in-depth comprehensive evaluation will be performed during the life of the project by the BNF panel of scientists who will review the progress made to achieve the established goals. The first evaluation will be scheduled during the third year of the project. Additional in-depth evaluations will be held during the sixth and ninth year of the project. The Evaluation factors will include the following: project achievements in addressing the needs of small farmers and rural women; LDC adoption of resultant technologies and products; net increases in crop production resulting from the use of BNF methodologies; and the achievement of project purposes and goals.

These evaluations will be based on monitoring reports, inspection of physical facilities, UH progress reports, technical publications, impact analysis reports and reports on participating countries involvement in the biological nitrogen fixation program. The evaluations will determine if project inputs are being provided as planned, that conditions and covenants of project workscope are being met, and that project outputs are being accomplished as planned. These evaluations will be the basis for recommended changes in the workscope necessary to achieve project purpose and end of project status.

7.4 Economic Assessment - The economic assessment activities will require the measurement of both primary and secondary benefits for the purpose of benefit/cost analysis. The resources available to this project will be allocated among competing uses according to perceived costs and benefits. The costs and benefits will be identified. Because this particular project has various components of the technology generation and transfer activities (scientific and service components), all costs (project costs and other contributions to the project) will be allocated at the outset of the project among appropriate cost categories by all participating cooperators. A complete cost accounting of project activities by program areas, will be undertaken to enable appropriate costs to be combined with various impact measures as they unfold during the life of the project and subsequent to the life of the project.

The cost accounting approach will annualize costs of facilities, equipment, supplies (expendables) and human resources (including travel) contributed to the project from all sources. Training (both short-term and long-term) is a significant component of the project that poses unique measurement problems for costs and benefits. A detailed periodic follow-up of all participants will be made. The participant follow-up will provide valuable insight into both the private and social costs/benefits of human capital investments. Data collected over time can help assess the "multiplier effect" of human capital investment relative to the more traditional tangible capital estimates.

Direct impact will be the increased yield and/or reduced necessity of nitrogenous fertilizer purchases. This impact will be based on yield performance observed from experimental field trials and on-farm trials that focus on the use of BNF technologies in farmers' systems. Where appropriate, macro economic issues will be considered - reduced foreign exchange commitment to fertilizer purchases is an example. In addition, efforts will be made to quantify the benefits associated with advances in BNF research and applied technologies.

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8.0 Project Conformity with Various Strategies and Guidelines

8.1 Project Conformity with the Strategy for Food and Agriculture Development Assistance

The objective of A.I.D.'s Food and Agricultural Policy and Strategy is to enable LDCs to become self-reliant in food, assure food security and contribute to broadly-based economic growth. This project is consistent with these objectives and the four elements of the policy listed below:

8.1.1 Improving Country Policies

This project is designed to improve the standard of living in agricultural areas by increasing agricultural production and the availability of food at a minimum cost. It will provide the LDCs with the ability to develop and improve agricultural policies, institutions, technologies and manpower resources needed to decrease the use of imported fertilizers and establish improved production systems using legumes and rhizobia to increase the nutrient level of soils.that will lead to increased agricultural production.

8.1.2 Providing Food Aid and Food Security

Increased production of food is necessary in most developing countries to reduce the need for food aid and increase food security. This project will enable LDCs to increase yields of food crops while reducing their dependence on expensive, imported fertilizer. It will also enhance the fertility of their soils, the natural resource base for sustained yields in the future.

8.1.3 Expanding the Role of the Private Sector

This project will provide for private sector involvement primarily under Program 5, which must be funded by the requesting missions, regional bureaus and/or LDC governments and institutions. However, it should be noted that private sector involvement is critical to the successful utilization of BNF technologies in the LDCs and must be promoted. In developed countries inoculum technology is implemented widely on a commercial scale, and demands for this technology continue to increase. Small private enterprises could easily be established in certain LDCs to provide high quality inoculum for marketing and sale to individual consumers, in-country programs, and government seed producers.

8.1.4 Development of Human Resources and Institutional Capacity

This project will strengthen human and institutional resources in the LDCs through academic, technical and on-the-job training, information transfer, and development of research, extension, and distribution networks.

8.2 Conformity with S&T's Ribbon Project Approach

This project will continue the ribbon approach established under the previous contracts with UH to link with and support the efforts of a range of bilateral, regional and global projects related to soil fertility requirements. Networks are one means for professionals to share technical information and to transfer research results from one region to another. However, networks among LDC professionals will not likely develop without a special catalyst (e.g., this project) to initiate the necessary action and provide the necessary funding.

It is intended that this project will coordinate and backstop common theme networks through workshops and the dissemination of scientific information. Some of the benefits of fostering the ribbon approach are as follows:

- Sharing of expertise among several aid donors and international institutions (e.g., FAO, UNDP, IRRI, and CIAT) promises in the long term to reduce the LDCs' dependency on any one donor for assistance.

- By pooling the resources of scientific organizations and personnel, each will have access to a much greater level of interdisciplinary expertise. This is especially beneficial to scientists and experts in the smaller and poorer LDCs. The results in one country will be effectively applied to other countries of similar environments.

8.3 Conformity with Regional Bureau Priorities

The agricultural research priority of the regional bureaus is to increase crop production. This project is designed to increase crop production by the use of BNF methods. Further, it supports and complements the objectives of improving agricultural production and supply systems in less favorable environments and encouraging minimum-purchased-input systems. It also strengthens LDC institutional capability by providing technical assistance and academic, technical and on-the-job training at UH and LDC national, regional and international institutions.

8.4 Conformity with the CPSS

The S&T/AGR portfolio is designed and managed to contribute to resolution of the two priority AID development problems most closely related to agricultural development, economic growth and hunger. This project, if successfully implemented in the LDCs, is consistent with both priorities. As stated above, this project is designed to address the four designated AID programmatic elements: technology development and transfer, institution-building, and private sector development. It is ranked one of the highest projects on the S&T/AGR priority listing.

8.5 Relationship to AID funded projects

To better use available technology, UH will investigate the possibility of collaboration with contractors, grantees and participating agencies which have contracts and agreements with S&T/AGR, missions and regional bureaus.

8.5.1 Office of Agriculture Projects

1) Collaborative Research Support Programs to increase crop production by the use of BNF methods:

   - Beans and Cowpeas
   - Peanuts

2) International agricultural research centers to increase crop production by the use of BNF methods.

   - IRRI for rice
   - IITA for cropping systems, including alley cropping in subhumid tropical areas of Africa
   - CIAT for common beans
   - IBPGR for collection, conservation and utilization of plant genetic resource materials

3) Other S&T/AGR Projects

   - Bio-technology Limiting Factors to test inoculant methodologies

   - Soybean Utilization and Research to conduct rhizobium inoculation experiments through the tropics.

   - International Benchmark Sites Network to integrate the scientific knowledge of agricultural production through computer modelling and selecting sites for the Environmental Constraints Program.

   - Agricultural Policy Analysis to improve knowledge and understanding of LDC decision-makers about key policy issues and constraints affecting the use of BNF.

   - Farming Systems Support to integrate the use of BNF into the farming systems used in the LDC.

   - Tissue Culture: The Genetic Technologies program calls for close collaboration with the Tissue Culture project being implemented by Colorado State University.

8.5.2 Other S&T Projects

It is anticipated that research on the topics cited below will be further developed and that NifTAL will be able to contribute to these efforts.

- Woody species
- Alley Cropping

8.6 Other AID Funded Projects

1) Office of the Science Advisor (SCI)

SCI, under its Program in Science and Technology Cooperation (PSTC), funds and administers a research grants program in areas of "research modules". plant biotechnology; and biomass resources and conversion technology. This PSTC program and the Improved BNF thru Biotechnology project can be mutually reinforcing. The exploratory grants funded by PSTC on N fixing legumes, which are limited in scope, can identify new areas for research under the Improved BNF thru Biotechnology project and the LDC scientists for involvement in the project. Conversely, the UH could investigate follow-on activities for PSTC to fund which deal with the N-fixing microorganisms.

SCI also funds a grants program administered by the Board on Science and Technology in Development (BOSTID) of the National Research Council of the National Academy of Sciences. The BNF aspects of grants supported under this project will complement the Improved BNF thru Biotechnology project. For example, research on BNF in fast-growing tree legumes, azolla-anabaena systems and nonsymbiotic, N-fixing associations funded by BOSTID can be beneficial to UH in its research activities implemented by NifTAL.

2) Mission funded production projects

- All production projects funded by the missions and regional bureaus will benefit from the BNF methodologies developed under the previous contracts and to be developed under this new project. The countries and activities are listed by Bureaus on pages 25, 26, and 27.

8.6 Other Programs

8.6.1 The USDA/ARS ongoing biotechnology research is directed toward the solution of high priority agricultural problems of national scope. These include reducing high costs of farm production and improving production efficiency. In the area of nitrogen fixation, ARS has a research program underway to genetically improve rhizobium bacteria to enhance nitrogen-fixation capacity of legume crops and transfer nitrogen-fixation genes to non-leguminous crops that now lack this capacity.

8.6.2 The United Nations Development Programme (UNDP) has sponsored research at the International Rice Research Institute (IRRI) since 1976 to investigate the potential of N_2-fixing system in lowland rice. This program seeks to ensure satisfactory rice yields while minimizing the use of chemical nitrogen fertilizer.

8.6.3 The Government of Mexico has signed a Declaration of Intent to collaborate with the University of Hawaii in BNF programs.

For additional information on programs working collaboratively with NifTAL, please refer to Appendix D.

9.0 Project Analysis

9.1 Economic Analysis

While direct benefits of research can not be observed before research results are obtained and put into use, some potential benefits can be mentioned. Investments in research activities have historically yielded high rates of return and the potential benefits from research projects are substantial. The purpose of agricultural research, in general, is to increase agricultural productivity with the smallest investment of resources. This increase occurs in several ways: 1) by increasing output or lowering costs (through substitution of less expensive resources), 2) by improving quality of farm products, 3) by the development of new products; and 4) by reducing the risk factor.

Direct cost-benefit studies of the agricultural research found an annual rate of return of 90 percent to investment in wheat research in Mexico, 77 percent to cotton in Brazil, 35-40 percent to maize in Peru, 75 percent to rice in Japan, and 60 percent to sugar cane research in India. In like fashion, high rates of return are expected to be associated with BNF research investments. Previous BNF research in developing countries showed evidence of considerable potential. Legume inoculation showed an average yield increase of 24 percent for 12 countries. However there was considerable variation among countries ranging from no yield response to a yield increase of approximately 80 percent.

A specific objective of this project is to reduce the dependence of developing countries on foreign powers for expensive fertilizer to produce basic foods for their peoples. In many LDCs the cost of fertilizer is already prohibitive, or not available. Many small farmers are unable to obtain fertilizer. However, biological methods can be used to improve soil fertility and increase crop production. With the development of good management practices, the use of BNF technologies, and a good information (extension) program, farmers will benefit by increasing crop production, and, if prices remain constant (or rise), higher farm income. Private sector involvement of production and distribution of legume inoculum will create investment opportunities; e.g., more jobs and increased income.

Another benefit to the developing countries by the use of BNF to increase agricultural production, is a decrease in fertilizer imports. In 1983/84, the developing countries (in aggregate) imported 6.95 million metric tons of fertilizer nitrogenous valued in excess of $3.6 billion. The level for 1985 is expected to be the same. If a sufficient number of farmers adopted BNF techniques to reduce imports by only 5 percent, a savings would be realized of $152.9 million annually. These savings could be extremely important to developing countries that are experiencing balance of payment problems. A reduction in fertilizer imports would result in savings of scarce foreign exchange that could be directed towards the priority problems of economic growth and hunger.

The benefits of agricultural research tend to affect other sectors of the economy, and indeed the world; e.g., consumers have abundant food supplies at relatively low costs; research techniques have applicability in other countries; and new markets and employment opportunities are created.

9.2 Social Soundness Analysis

This project provides strategies for the development and promotion of agrobiotechnologies. The development of such technology can have substantial social impact on the economies and environments of the LDCs.

9.2.1 Beneficiaries

Several groups will be beneficiaries of this project: 1) LDC scientists will benefit directly from training in the latest biotechnologies; 2) their institutions will gain an enhanced ability to establish programs in these areas; 3) farmers will have more crops to sell and an increase in income; 4) consumers will have more food; and 5) small business entrepreneurs will be able to use the biotechnology developed at UH to develop new product lines and new markets.

Participation in the research networks that form the core of Program 1 - Genetic Technologies, Program 2 - Development of Methodologies, and Program 3 - Environmental Data Collection will strengthen the research capability of LDC institutions and allow them to obtain information and management tools, such as the predictive model generated by Program 3, at a minimum expense. Long-term collaboration of U.S. and developing country researchers is an expected spin-off of Program 3. In addition, other networks may arise as a result of the positive experience of participants in the research. Such a ribbon effect of networking has been the experience of the NifTAL project with the INLIT program.

Program 5 will provide small business entrepreneurs with technical information on the production and distribution of rhizobium inoculant to start small biotech rhizobium inoculant businesses. Not only will the individuals have access to NifTAL expertise, but through small workshops will be held to address the problems of inoculant production. The concept of production networks of inoculant entrepreneurs is perhaps novel, but likely to bear fruit if the individuals involved see benefit in collectively solving common problems. AID missions and other foreign donors will benefit from the presence of Regional Resource Centers in their areas. These Centers can not only provide technical expertise and assist in mission project development, but through their extensive network of collaborators assist in the identification of individuals or organizations that could participate in development projects.

Small LDC farmers will also benefit directly through basic research conducted by LDC scientists and institutions and the activities of the small business group. The activities of the latter, in particular, will assure the availability of high quality inoculants for legume crops and trees which will improve production and lead to increased income and well being for the small farmers. The number of farmers who benefit will depend upon the success of BNF programs in a particular developing country. Successful implementation of BNF technology through the proposed promotional programs will lead to increased output and a rise in the standard of living for small farmers.

9.2.2 Social Impact

The impact of this project is likely to go far beyond the small farmers. Adoption of biotechnology is likely to have a favorable impact on the balance of payments of the participating LDCs. In the case of BNF technologies, less nitrogen will have to be imported. This will free financial resources from the purchase of fertilizer to be used in improving living conditions; e.g., improved roads, water systems, schools, and health facilities. The development of small inoculum industries would provide employment for a small number of people in the participating LDC.

9.2.3 Women in Development

Women in developing countries will participate and contribute substantially to all levels of the BNF program. As women farmers grow at least 50 percent of the world's food (in parts of Africa over 90 percent), they will be the direct beneficiaries of the improved methods and technologies developed and the resultant cost savings. By using BNF technologies, LDC farm women will be doing different work, but will benefit in terms of higher yields, especially when faced with fertilizer shortages. Women farmers will be included in the field tests and later extension work. Since they comprise the core of farm workers in developing countries, their education is a critical component of this project.

Additionally, the resources of women in research science in the developing countries will be drawn upon and broadened through this program. The participation of women in the scientific community varies from country to country. In the International Agricultural Research Centers (IARCs), approximately 5.6 percent of the senior researchers are female. The LDC women in science will contribute by participating in this new program as research scientists, laboratory managers, and technicians. They will contribute their scientific expertise and technical skill as well as their clear understanding of their very specific country problems. The program in turn will aid in the development and growth of the pool of LDC women in science through the various exchange and training aspects of the program components.

Women have occupied key positions under the previous project; e.g., Dr. Joann Roskoski, is the senior tropical plant ecologist and plant physiologist holding a key position in the project. Over 25 percent of the participants, both at the technical level and M.S. and Ph.D. levels have been trained under the previous project. They will continue to be selected for the various training programs under the new project.

9.3 Administrative Feasibility

The University of Hawaii, which successfully administered the previous BNF project, is highly qualified to manage this new project. The University has assembled for this project a world renowned staff of scientists from interrelated disciplines, who were carefully selected on the basis of their ability and experience in tropical agriculture in the LDCs. The University is experienced in coordinating activities in the LDCs which was a major component of the previous project. The primary and basic focus of this Hawaiian staff is on the biological fixing of nitrogen in depleted soils of the tropics.

9.4 Technical Analysis

When BNF is first introduced into an area there is a dramatic increase in yield. In certain areas, a five-fold increase has been obtained with inoculation of certain legumes. With continued use and adaptation of BNF technology this increase reaches a plateau level. The technical aspects of this project are described in Appendices D (State-of-Art) and E (Technical Review). The scope of work in this new project is technically feasible, and is especially attractive because national scientists and LDC entrepreneurs will be involved directly in the activities in the programs; i.e., Program 3.1.1 - Genetic Technologies; Program 3.1.2 - Monitoring Microorganisms; Program 3.1.3 - Environmental Data Collection; 3.1.4 - Regional Resource Centers; and 3.1.5 - Inoculant Production Technical Assistance.

9.5 Environmental Constraints

This project is designed to increase crop productivity and to reduce dependency on agricultural imports. It provides basic and adaptive research, training, technology transfer and networking activities to strengthen human resources and institutional capacity to develop and implement economically

efficient, socially acceptable and environmentally sound programs which will use biological nitrogen fixation to increase crop productivity. To the extent that the project involves the use of chemically treated germ plasm, the grantee will establish that none of the activities will involve the use of pesticides that fall in the restricted category in the U.S. or have been banned, suspended or withdrawn from use for agricultural purposes in the U.S.

In all other respects, the initial phase of the project will have minimal or no direct effect on the environment and is exempt from the Environmental Procedures of 22 CFR 216.2 for the reasons described in 22 CFR 216.2(c) (i), (iii), (2), (i), (iii), (v) and (xiv). A negative determination is recommended. However, as research progresses, it is expected that new microorganisms will be developed. It is difficult at this time to assess the impact of these microorganisms since their exact nature is unknown. The field testing of such products after development will follow all appropriate Federal and international regulations. The anticipated impact of these products when ready for field testing will be evaluated on a case-by-case basis.

The Environmental Threshold Determination and Initial Environmental Examination are attached as Appendix J.

9.6 Financial Analysis and Funding of the New Project

9.6.1 Financial Analysis

The Input-Output budgets on pages 22 and 23 and the proposed staffing shown on pages 24 are realistic for the level of effort and time frame proposed in this new project. As indicated above, we will expect participating missions, regional bureaus and LDC institutions to provide financial and managerial support to the project; especially for those activities under Programs 3.1.4 and 3.1.5. In addition, scientists and technicians will be trained primarily under the missions participant training programs and by LDC institutions.

9.6.2 Funding for New Project

S&T/AGR is requesting approval for a ten-year project at a level of $19.99 million, which includes an S&T/AGR core budget of $9.69 million, and an expected $10.30 million contribution from the missions, regional bureaus and other AID/W offices to be funded by Delivery Orders under the Basic Ordering Agreement. The participating LDC institutions must cover the local costs of the Regional BNF Resource Centers. In addition, the University of Hawaii is expected to contribute $2.468 million to achieve the goal and purpose of this project. These cost estimates are based on the calculated level of effort needed; i.e., personnel, travel and allowances, equipment, supplies and services and other requirements necessary to achieve targeted outputs of the types and magnitude as set forth in the logical framework (Appendix A).

4/18/86:WANG BNF2
Revised:4/30/86:Revised:5/7/86

DIMPEX

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX A - PROJECT DESIGN SUMMARY - LOGICAL FRAMEWORK

Revised:4/29/86: Revised 5/8/86
DIMPEX

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AID 1020-20 (1-72)

PROJECT DESIGN SUMMARY
LOGICAL FRAMEWORK

Life of Project:
From FY 86 to FY 96
Total U.S. Funding
Date Prepared: April 21, 1986
Revised April 29, 1986

Project Title & Number: Improved BNF through Biotech - 936-4177

NARRATIVE SUMMARY	OBJECTIVELY VERIFIABLE INDICATORS	MEANS OF VERIFICATION	IMPORTANT ASSUMPTIONS
Program or Sector Goal: The broader objective to which this project contributes: To: 1) increase crop productivity; 2) reduce inefficient use of imported agricultural inputs; and 3) encourage sustainable agricultural production systems.	Measures of Goal Achievement: - Overall increase in crop productivity. - Efficient use of expensive imported agricultural inputs; e.g. fertilizer. - Sustainable agricultural production systems in place.	- National Surveys. - AID/W and Reports. - FAO production Statistics - Impact Evaluation - FAO Fertilizer import/export data.	Assumptions for achieving goal targets: - Food production remains high priority in LDC's. - LDCs will use BNF. - Environmental conditions are conducive to increasing crop production.
Project Purpose: The purpose of this project is to. 1) increase the efficiency of nitrogen fixing micro-organisms adapted to LDC conditions thru methods of biotechnology; 2) promote the use of BNF in LDCs by assisting them to adapt, use and disseminate information about BNF; and 3) increase their capacity to produce and distribute BNF inocula.	Conditions that will indicate purpose has been achieved: End of project status. 1. Improved BNF materials and techniques adapted and utilized at farm level in LDCs. 2. Increased use of legumes in multiple purpose roles. 3. Decrease in use of nitrogen fertilizer for legumes. 4. Legume inoculum production plants established in LDCs. 5. Trained LDC scientists.	1,2,3,4. - Annual reports from Missions and grantee. - On-site visits. - Impact Evaluations. - LDC statistics and reports. - Market Reports. - FAO Fertilizer import/export data. 5. - Training reports and degrees given.	Assumptions for achieving purpose: - LDC scientists can acquire sufficient competence to conduct useful research within a three to nine year period. - LDC Farmers are willing to use new BNF Technologies. - LDC entrepreneurs are capable of establishing inoculum production plants.

AID 1020-25 (1-72)

PROJECT DESIGN SUMMARY
LOGICAL FRAMEWORK

Life of Project:
From FY 86 to FY 96
Total U.S. Funding
Date Prepared: April 21, 1986

Project Title & Number: Improved BNF through Biotech - 936-4177

NARRATIVE SUMMARY Outputs:	OBJECTIVELY VERIFIABLE INDICATORS Magnitude of Outputs:	MEANS OF VERIFICATION	IMPORTANT ASSUMPTIONS Assumptions for achieving outputs:
		Programs 3.1.1, 3.1.2, 3.1.3, 3.1.4, and 3.1.5. --UH Technical and Research Reports. --Annual Activity Reports. --Training Activities. --Impact Analysis Reports. --Trip Reports. --Evaluations. --Site Visits. --Mission and LDC Reports. --Trained LDC scientists in positions of responsibility.	Programs 3.1.1, 3.1.2, and 3.1.3. --Selected IARCs and LDC institutions and scientists will collaborate with NifTAL in training, research, networking and inoculum production. --LDC scientists are available for training and will be funded by Missions and LDCs. Program 3.1.1 --Climatic conditions on Maui continue to provide ideal environment for research and training on N-fixing microorganisms --Supercompetitor rhizobium genes can be identified. Program 3.1.2 --Microorganisms' antibodies produced at UH can be used effectively in LDCs --Teaching modules for monitoring methodologies are used in LDCs and IARCs.
Program 3.1.1 Genetic Technologies for Improvement of Rhizobium/Legume Symbiosis for Crops and Trees.	1) 1 resource bank of rhizobium 2) 6 genetically improved rhizobium strains 3) 5 improved host legumes 4) 2,000 germplasm collected, 100 tested, 2,000 catalogued, 4,000 distributed 5) *40 LDC scientists trained 6) 20 publications distributed.		
Program 3.1.2 Development of Methods for Monitoring Microorganisms.	1) 1 resource bank of antibodies 2) 100 resource germplasm of microorganisms characterized 3) 23 publications distributed 4) 5 scientists trained at NifTAL 5) Biotech 6-week training course held annually 6) 5 training manuals published and distributed 7) 250 collaborative linkage with LDC institutions established 8) 100 antisera and genetically marked organisms provided LDC scientists in 50 countries.		

* Numbers reflect person-year and may include the same person for more than one year.

PROJECT DESIGN SUMMARY
LOGICAL FRAMEWORK

Project Title & Number: Improved BNF through Biotech - 936-4177

Life of Project:
From FY 86 to FY 96
Total U.S. Funding
Date Prepared: April 21, 1986

Revised 4/29/86

NARRATIVE SUMMARY	OBJECTIVELY VERIFIABLE INDICATORS	MEANS OF VERIFICATION	IMPORTANT ASSUMPTIONS
Outputs: Continued	Magnitude of Outputs:	Programs 3.1.1, 3.1.2, 3.1.3, 3.1.4, and 3.1.5.	Assumptions for achieving outputs:
Program 3.1.3 Environmental Data Base to maximize performance of Biological Nitrogen Fixation.	1) 1 model created to predict inoculation response 2) 3 regional networks of LDC scientists 3) A linkages established among networks 4) Enhanced capacity of 100 LDC institutions to solve BNF problems 5) *41 LDC professionals trained 6) 20 publications distributed. 7) One short-course held annually	--UII Technical and Research Reports. --Annual Activity Reports. --Training Activities. --Impact Analysis Reports. --Trip Reports. --Evaluations. --Site Visits. --Mission and LDC Reports. --Trained LDC scientists in positions of responsibilities.	Program 3.1.3 --LDC institutions continue to improve their capacity to solve important agricultural problems --Model to predict inoculation response by tropical legumes will be used by the LDCs and IARCs.
Program 3.1.4 Regional Resource Centers.	1) 4 centers operational 2) 1 newsletter distributed annually from each center 3) NifTAL strains and materials used and fed back to UII 4) BNF Technologies incorporated in LDC projects 5) 60 training courses held 6) 1 to 2 scientists conducting research at each center 7) 18 scientific articles published in refereed journals.		Program 3.1.4 --Suitable LDC host institutions will participate in regional BNF Resource Centers --Regional activities will be encouraged --Funding of the Centers will come from Missions and Regional Bureaus.
Program 3.1.5 Commercial Inoculant technical assistance	1) 1 inoculum production plant established in each region 2) 2 private enterprises incorporated in each region - total 6 3) Inoculum produced and available for marketing 4) 5,000 "How to" publications distributed 5) 150 entrepreneurs trained. 6) Short courses and workshops held annually		Program 3.1.5 --LDCs interested in establishing inoculum production plants --Private sector promotional activities are encouraged --Sufficient market will exist to support local production --National economic policies foster private entrepreneurship --Small entrepreneurs can maintain consistent quality --Start up capital is available --Distribution infrastructure is adequate.

* Numbers reflect person-year and may include the same person for more than one year.

IMPROVED BIOLOGICAL NITROGEN FIXATION (BNF) THROUGH BIOTECHNOLOGY
TEN YEAR SUMMARY
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East			Grand	Grand
	Office of Agriculture Core Budget																	Total	Total
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Missions	Proj.
Salaries	1,510	1,110	1,230	600	600	5,050	1,000	6,050	820	-	820	930	-	930	1,700	-	1,700	3,450	9,500
Fringe(20%)*	300	220	250	120	120	1,010	210	1,220	248	-	248	299	-	299	534	-	534	1,081	2,301
Consultants	-	-	-	-	-	-	-	-	-	715	715	-	770	770	-	1,135	1,135	2,620	2,620
Travel	50	50	100	100	90	390	-	390	48	117	165	58	125	183	112	190	302	650	1,040
Publication	60	60	120	100	100	440	-	440	30	-	30	33	-	33	56	-	56	119	559
Supplies	100	100	100	100	160	560	-	560	72	164	236	74	171	245	130	140	270	751	1,311
Total	2,020	1,540	1,800	1,020	1,070	7,450	1,210	8,660	1,218	996	2,214	1,394	1,066	2,460	2,532	1,465	3,997	8,671	17,331
Indirect **	410	310	360	200	210	1,490	-	1,490	172	179	351	198	209	407	360	285	645	1,403	2,893
Equipment	110	40	40	-	-	190	1,258	1,448	-	-	-	-	-	-	-	-	-	-	1,448
Training	40	40	40	-	-	120	-	120	60	-	60	58	-	58	108	-	108	226	346
Utilities	100	150	100	-	-	350	-	350	-	-	-	-	-	-	-	-	-	-	350
Total	2,680	2,080	2,340	1,220	1,280	9,600	2,468	12,068	1,450	1,175	2,625	1,650	1,275	2,925	3,000	1,750	4,750	10,300	22,368
Evaluation						90	-0-	90	-	-	-	-	-	-	-	-	-	-	90
Grand Total	2,680	2,080	2,340	1,220	1,280	9,690	2,468	12,158	1,450	1,175	2,625	1,650	1,275	2,925	3,000	1,750	4,750	10,300	22,458

* Fringe Benefits for the scientists stationed at the Regional Resource Centers includes housing and other local benefits.
** Indirect cost for staff, travel, publications and supplies needed at UH is computed at 20% of these charges.
Indirect cost for staff, travel, publications and supplies needed for the Regional Centers is computed at 14% of these charges.

WANG 1161C
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Appendix A
Logframe
Page 4 of 8

IMPROVED BIOLOGICAL NITROGEN FIXATION (BNF) THROUGH BIOTECHNOLOGY

FIRST FIVE YEARS
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East				
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Grand Total Missions	Grand Total Proj.
Salaries	755	555	615	300	300	2,525	500	3,025	420	-	420	700	-	700	1,280	-	1,280	2,400	5,425
Fringe(20%)*	150	110	125	60	60	505	105	610	135	-	135	225	-	225	399	-	399	759	1,369
Consultants	-	-	-	-	-	-	-	-	-	285	285	-	330	330	-	675	675	1,290	1,290
Travel	25	25	50	50	45	195	-	195	24	47	71	42	55	97	88	120	208	376	571
Publication	30	30	60	50	50	220	-	220	15	-	15	25	-	25	41	-	41	81	301
Supplies	50	50	50	50	80	280	-	280	36	66	102	58	77	135	94	45	139	376	656
Total	1,010	770	900	510	535	3,725	605	4,330	630	398	1,028	1,050	462	1,512	1,902	840	2,742	5,282	9,612
Indirect **	205	155	180	100	105	745	-	745	90	77	167	150	88	238	270	160	430	835	1,580
Equipment	55	20	20	-	-	95	629	724	-	-	-	-	-	-	-	-	-	-	724
Training	20	20	20	-	-	60	-	60	30	-	30	50	-	50	78	-	78	158	218
Utilities	50	75	50	-	-	175	-	175	-	-	-	-	-	-	-	-	-	-	175
Grand Total	1,340	1,040	1,170	610	640	4,800	1,234	6,034	750	475	1,225	1,250	550	1,800	2,250	1,000	3,250	6,275	12,309
Evaluation						30		30											30
Grand Total	1,340	1,040	1,170	610	640	4,830	1,234	6,064	750	475	1,225	1,250	550	1,800	2,250	1,000	3,250	6,275	12,339

SECOND FIVE YEARS
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East				
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Grand Total Missions	Grand Total Proj.
Salaries	755	555	615	300	300	2,525	500	3,025	400	-	400	230	-	230	420	-	420	1,050	4,075
Fringe(20%)*	150	110	125	60	60	505	105	610	113	-	113	74	-	74	135	-	135	322	932
Consultants	-	-	-	-	-	-	-	-	-	430	430	-	440	440	-	460	460	1,330	1,330
Travel	25	25	50	50	45	195	-	195	24	70	94	16	70	86	24	70	94	274	469
Publication	30	30	60	55	50	220	-	220	15	-	15	8	-	8	15	-	15	38	258
Supplies	50	50	50	50	80	280	-	280	36	98	134	16	94	110	36	95	131	375	655
Total	1,010	770	900	510	535	3,725	605	4,330	588	598	1,186	344	604	948	630	625	1,255	3,389	7,719
Indirect **	205	155	180	100	105	745	-	745	82	102	184	48	121	169	90	125	215	568	1,313
Equipment	55	20	20	-	-	95	629	724	-	-	-	-	-	-	-	-	-	-	724
Training	20	20	20	-	-	60	-	60	30	-	30	8	-	8	30	-	30	68	128
Utilities	50	75	50	-	-	175	-	175	-	-	-	-	-	-	-	-	-	-	175
Grand Total	1,340	1,040	1,170	610	640	4,800	1,234	6,034	700	700	1,400	400	725	1,125	750	750	1,500	4,025	10,059
Evaluation						60		60											60
Grand Total	1,340	1,040	1,170	610	640	4,860	1,234	6,094	700	700	1,400	400	725	1,125	750	750	1,500	4,025	10,119

* Fringe Benefits for the scientists stationed at the Regional Resource Centers includes housing and other local benefits.
** Indirect cost for staff, travel, publications and supplies needed at UH is computed at 20% of these charges.
Indirect cost for staff, travel, publications and supplies needed for the Regional Centers is computed at 14% of these charges.

WANG 1161C Revised 4/29/86

IMPROVED BIOLOGICAL NITROGEN FIXATION (BNF) THROUGH BIOTECHNOLOGY

FY 86
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East				
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Grand Total Missions	Grand Total Proj.
Salaries	32	23	26	12	12	105	-	105											105
Fringe(20%)*	6	5	5	2	2	20	-	20											20
Consultants	-	-	-	-	-	-	-	-											-
Travel	-	-	-	-	-	-	-	-											-
Publication	3	3	3	4	4	17	-	17											17
Supplies	5	5	5	5	6	26	-	26											26
Total	46	36	39	23	24	168	0	168											168
Indirect **	9	7	7	4	5	32	-	32											32
Equipment	-	-	-	-	-	-	-	-											-
Training	-	-	-	-	-	-	-	-											-
Utilities	-	-	-	-	-	-	-	-											-
Grand Total	55	43	46	27	29	200	0	200											200

FY 87
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East				
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Grand Total Missions	Grand Total Proj.
Salaries	151	111	123	60	60	505	100	605	-	-	-	140	-	140	280	-	280	420	1,025
Fringe(20%)*	30	22	25	12	12	101	21	122	-	-	-	45	-	45	90	-	90	135	257
Consultants	-	-	-	-	-	-	-	-	-	30	30	-	30	30	-	134	134	194	194
Travel	5	5	10	10	9	39	-	39	-	5	5	10	5	15	20	24	44	64	103
Publication	6	6	12	10	10	44	-	44	-	-	-	5	-	5	10	-	10	15	59
Supplies	10	10	10	10	16	56	-	56	-	7	7	10	7	17	20	10	30	54	110
Total	202	154	180	102	107	745	121	866	0	42	42	210	42	252	420	168	588	882	1,748
Indirect **	41	31	36	20	21	149	-	149	-	8	8	30	8	38	60	32	92	138	287
Equipment	11	4	4	-	-	19	184	203	-	-	-	-	-	-	-	-	-	-	203
Training	4	4	4	-	-	12	-	12	-	-	-	10	-	10	20	0	20	30	42
Utilities	10	15	10	-	-	35	-	35	-	-	-	-	-	-	-	-	-	-	35
Grand Total	268	208	234	122	128	960	305	1,265	0	50	50	250	50	300	500	200	700	1,050	2,315

Fringe Benefits for the scientists stationed at the Regional Resource Centers includes housing and other local benefits.
Indirect cost for staff, travel, publications and supplies needed at UH is computed at 20% of these charges.
Indirect cost for staff, travel, publications and supplies needed for the Regional Centers is computed at 14% of these charges.

WANG 1161C

Revised 4/29/86

IMPROVED BIOLOGICAL NITROGEN FIXATION (BNF) THROUGH BIOTECHNOLOGY

FY 88
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East				
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Grand Total Missions	Grand Total Proj.
Salaries	151	111	123	60	60	505	100	605	-	-	-	140	-	140	280	-	280	420	1,025
Fringe(20%)*	30	22	25	12	12	101	21	122	-	-	-	45	-	45	90	-	90	135	257
Consultants	-	-	-	-	-	-	-	-	-	45	45	-	60	60	-	134	134	239	239
Travel	5	5	10	10	9	39	-	39	-	7	7	8	10	18	20	24	44	69	108
Publication	6	6	12	10	10	44	-	44	-	-	-	5	-	5	10	-	10	15	59
Supplies	10	10	10	10	16	56	-	56	-	10	10	12	14	26	20	10	30	66	122
Total	202	154	180	102	107	745	121	866	0	62	62	210	84	294	420	168	588	944	1,810
Indirect **	41	31	36	20	21	149	-	149	-	13	13	30	16	46	60	32	92	151	300
Equipment	11	4	4	-	-	19	217	236	-	-	-	-	-	-	-	-	-	-	236
Training	4	4	4	-	-	12	-	12	-	-	-	10	-	10	20	-	20	30	42
Utilities	10	15	10	-	-	35	-	35	-	-	-	-	-	-	-	-	-	-	35
Grand Total	268	208	234	122	128	960	338	1,298	0	75	75	250	100	350	500	200	700	1,125	2,423

FY 89
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East				
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Grand Total Missions	Grand Total Proj.
Salaries	151	111	123	60	60	505	100	605	140	-	140	140	-	140	280	-	280	560	1,165
Fringe(20%)*	30	22	25	12	12	101	21	122	45	-	45	45	-	45	90	-	90	180	302
Consultants	-	-	-	-	-	-	-	-	-	60	60	-	60	60	-	134	134	254	254
Travel	5	5	10	10	9	39	-	39	8	10	18	8	10	18	20	24	44	80	119
Publication	6	6	12	10	10	44	-	44	5	-	5	5	-	5	10	-	10	20	64
Supplies	10	10	10	10	16	56	-	56	12	14	26	12	14	26	20	10	30	82	138
Total	202	154	180	102	107	745	121	866	210	84	294	210	84	294	420	168	588	1,176	2,042
Indirect **	41	31	36	20	21	149	-	149	30	16	46	30	16	46	60	32	92	184	333
Equipment	11	4	4	-	-	19	85	104	-	-	-	-	-	-	-	-	-	-	104
Training	4	4	4	-	-	12	-	12	10	-	10	10	-	10	20	-	20	40	52
Utilities	10	15	10	-	-	35	-	35	-	-	-	-	-	-	-	-	-	-	35
Grand Total	268	208	234	122	128	960	206	1,166	250	100	350	250	100	350	500	200	700	1,400	2,566
Evaluation						30		30											30
Grand Total	268	208	234	122	128	990	206	1,196	250	100	350	250	100	350	500	200	700	1,400	2,596

* Fringe Benefits for the scientists stationed at the Regional Resource Centers includes housing and other local benefits.
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Indirect cost for staff, travel, publications and supplies needed for the Regional Centers is computed at 14% of these charges.

Revised 4/29/86

Appendix A
Logframe
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IMPROVED BIOLOGICAL NITROGEN FIXATION (BNF) THROUGH BIOTECHNOLOGY

FY 90
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East			Grand	Grand
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Total Missions	Total Proj.
Salaries	151	111	123	60	60	505	100	605	140	-	140	140	-	140	230	-	230	510	1,115
Fringe(20%)*	30	22	25	12	12	101	21	122	45	-	45	45	-	45	74	-	74	164	286
Consultants	-	-	-	-	-	-	-	-	-	60	60	-	90	90	-	134	134	284	284
Travel	5	5	10	10	9	39	-	39	8	10	18	8	15	23	16	24	40	81	120
Publication	6	6	12	10	10	44	-	44	5	-	5	5	-	5	8	-	8	18	62
Supplies	10	10	10	10	16	56	-	56	12	14	26	12	21	33	16	10	26	85	141
Total	202	154	180	102	107	745	121	866	210	84	294	210	126	336	344	168	512	1,142	2,008
Indirect **	41	31	36	20	21	149	-	149	30	16	46	30	24	54	48	32	80	180	329
Equipment	11	4	4	-	-	19	93	112	-	-	-	-	-	-	-	-	-	-	112
Training	4	4	4	-	-	12	-	12	10	-	10	10	-	10	8	-	8	28	40
Utilities	10	15	10	-	-	35	-	35	-	-	-	-	-	-	-	-	-	-	35
Grand Total	268	208	234	122	128	960	214	1,174	250	100	350	250	150	400	400	200	600	1,350	2,524

FY 91
(In thousands)

	Cooperative Agreement								Latin America and Caribbean			Africa			Asia and Near East			Grand	Grand
	Office of Agriculture Core Budget																		
	Prog. No.1	Prog. No.2	Prog. No.3	Prog. No.4	Prog. No.5	Grand Total	Univ. Hawaii	Total C.A.	Region Center	Short Term	Total LAC	Region Center	Short Term	Total AFR	Region Center	Short Term	Total ANE	Total Missions	Total Proj.
Salaries	119	88	97	48	48	400	100	500	140	-	140	140	-	140	210	-	210	490	990
Fringe(20%)*	24	17	20	10	10	81	21	102	45	-	45	45	-	45	55	-	55	145	247
Consultants	-	-	-	-	-	-	-	-	-	90	90	-	90	90	-	139	139	319	319
Travel	5	5	10	10	9	39	-	39	8	15	23	8	15	23	12	24	36	82	121
Publication	3	3	9	6	6	27	-	27	5	-	5	5	-	5	3	-	3	13	40
Supplies	5	5	5	5	10	30	-	30	12	21	33	12	21	33	18	5	23	89	119
Total	156	118	141	79	83	577	121	698	210	126	336	210	126	336	298	168	466	1,138	1,836
Indirect **	32	24	29	16	16	117	-	117	30	24	54	30	24	54	42	32	74	182	299
Equipment	11	4	4	-	-	19	50	69	-	-	-	-	-	-	-	-	-	-	69
Training	4	4	4	-	-	12	-	12	10	-	10	10	-	10	10	-	10	30	42
Utilities	10	15	10	-	-	35	-	35	-	-	-	-	-	-	-	-	-	-	35
Grand Total	213	165	188	95	99	760	171	931	250	150	400	250	150	400	350	200	550	1,350	2,281

* Fringe Benefits for the scientists stationed at the Regional Resource Centers includes housing and other local benefits.
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Indirect cost for staff, travel, publications and supplies needed for the Regional Centers is computed at 14% of these charges.

WANG 1161C

Revised 4/29/86

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX B - CRITICAL PERFORMANCE INDICATORS

Revised:4/29/86: Revised 5/8/86
DIMPEX

IMPROVED BNF THRU BIOTECH
CRITICAL PERFORMANCE INDICATORS

	Year One	Year Two	Year Three	Year Four	Year Five	Year Six	Year Seven	Year Eight	Year Nine	Year Ten	TOTAL
Program 3.1.1 Genetic Technologies											
Resource Rhizobium Bank	1	-	-	-	-	-	-	-	-	-	1
Genetically Improved Rhizobium Strains	-	1	-	2	-	2	-	1	-	-	6
Improved Host Legumes	-	-	1	-	-	2	-	-	2	-	5
Germplasm Collected	1,700	50	50	50	50	50	50	-	-	-	2,000
Germplasm Tested	10	10	10	10	10	10	10	10	10	10	100
Germplasm Catalogued	-	1,700	50	50	50	50	50	50	-	-	2,000
Germplasm Distributed	400	400	400	400	400	400	400	400	400	400	4,000
LDC Scientists Trained	-	4	4	5	5	5	5	4	4	4	40
Publications Distributed	2	2	2	2	2	2	2	2	2	2	20
Program 3.1.2 Development of Methods											
One Resource Bank of Antibodies	1	-	-	-	-	-	-	-	-	-	1
Resource Germplasm of Microorganisms Characterized	10	10	10	10	10	10	10	10	10	10	100
Publications Distributed	1	2	3	3	3	3	2	2	2	2	23
Five Scientists Trained at NIFTAL	-	1	1	1	1	1	-	-	-	-	5
Six-week Biotech Training Courses Held Annually	1	1	1	1	1	1	1	1	1	1	10
Training Manuals Published & Distributed	1	-	1	1	-	1	1	-	-	-	5
Collaborative Linkages w/LDC Institution Established	20	30	50	-	50	-	50	-	50	-	250
Antisera, Antibodies & Genetically marked Organisms Provided LDC Scientists in 50 Countries	10	20	30	20	10	10	-	-	-	-	100
Program 3.1.3 Environmental Data Base											
One Model Created to Predict Inoculation Response	-	-	-	-	1	-	-	-	-	-	1
Three Regional Networks of LDC Scientists	-	1	1	1	-	-	-	-	-	-	3
Linkages Established Among Networks	x	x	x	x	x	x	x	x	x	x	
Enhanced Capacity of 100 LDC Institutions to Solve BNF Problems	-	10	10	20	30	10	10	10	-	-	100
LDC Professionals Trained at MS and Ph.D. Levels	-	4	4	5	5	6	5	4	4	4	41
Publications Distributed	-	2	2	2	2	2	2	2	3	3	20

Revised 4/29/86

IMPROVED BNF THRU BIOTECH
CRITICAL PERFORMANCE INDICATORS

	Year One	Year Two	Year Three	Year Four	Year Five	Year Six	Year Seven	Year Eight	Year Nine	Year Ten	TOTAL
Program 3.1.4 Regional Resource Centers											
Four Centers Operational	3	–	1	–	–	–	–	–	–	–	4
One Newsletter Distributed Annually From Each Center	1	1	1	1	1	1	1	1	1	1	10
NifTAL Strains and Materials Used and Fed Back provided to UH	x	x	x	x	x	x	x	x	x	x	
BNF Technologies Incorporated in LDC Projects	x	x	x	x	x	x	x	x	x	x	
Training Courses Held	6	6	6	7	7	8	7	7	6	–	60
One to Two Scientists Conducting Research at Each Center	x	x	x	x	x	x	x	x	x	x	
Scientific Articles Published in Refereed Journals	1	1	2	2	2	2	2	2	2	2	18
Program 3.1.5 Commercial Inoculant Technical Assistance											
One Inoculum Production Plant Established in Each Region		–	1	–	–	1	–	1	–	–	3
Private Enterprises Incorporated in Each Region		2	–	2	–	2	–	–	–	–	6
Inoculum Produced and Available for Marketing		x	x	x	x	x	x	x	x	x	
"How to" Publications Distributed		x		x		x		x		x	
Entrepreneurs Trained		20	30	30	20	30	20	–	–	–	150

Revised:4/29/86

Appendix B
Page 2 of 2

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX C - NIFTAL CONNECTION TO SMALL SCALE FARMERS AND PARTICIPANTS

Revised:4/29/86: Revised 5/8/86
DIMPEX

It has been ten years since NifTAL began its work at the University of Hawaii under contract to USAID. Our goal was, and still is, to reduce dependence of small farmers on costly nitrogen fertilizers for production of high quality food. This is accomplished by doing research and providing assistance to *Rhizobium* and legume programs throughout the tropics to more effectively exploit the *Rhizobium*-legume symbiosis.

These have been productive years and, like a well balanced wheel, momentum has carried us forward and kept us moving ahead. Whether by offering innovative training courses or technical expertise in setting up inoculant production facilities or by providing services from a regional center, NifTAL is working toward meeting its goal.

TECHNOLOGY TRANSFER

The present worldwide emphasis on biotechnology development is well met in NifTAL's adaptation and promotion of the naturally occurring phenomenon of BNF. Dr. N. C. Brady, Senior Assistant Administrator for Science and Technology, USAID, has stated in a recent article on tranferring technology for small scale farming, ". . . technology worthy of adoption cannot be transferred (to farmers with limited resources) unless existing technology is modified, or new technology directly related to the developing countries is developed, or both." This is what NifTAL has done, is doing, and will continue to do.

INTEGRATED APPROACH

To achieve its overall project goal, NifTAL has used an integrated approach involving research, outreach, and training activities. Through research, a comprehensive collection of *Rhizobium* germplasm was developed containing effective strains adapted to tropical conditions which are distributed to researchers and inoculant producers in the tropics. Also developed have been improved techniques and systems for ensuring dependable effective nodulation and maximum utilization of legume-fixed nitrogen in cropping systems.

APPROPRIATE TECHNOLOGIES

NifTAL has developed appropriate and usable technologies for preparing much needed inoculants. Flask-sized to small scale, practical, low or moderate cost production systems have been improved or designed. NifTAL staff have aided in the implementation of inoculant production facilities in Bangladesh, Burma, Egypt, Sri Lanka, Thailand, and Zambia. We have drawn from the expertise of *Rhizobium* inoculant specialists—both in the U.S. and other countries. This expertise combined with the innovative efforts of NifTAL scientists has had excellent results.

COLLABORATIVE NETWORKING

NifTAL researchers realize the benefits of collaborative efforts and have used international contacts to make best use of networking through NifTAL's International Network of Legume Inoculation Trials (INLIT). More than 200 LDC scientists from 53 nations have participated in the INLIT program.

TRAINING METHODS

Over 200 linkages have been formed through NifTAL's Legume/*Rhizobium* Technology training courses. Nine 6-week training courses have been held. The first four were held in Hawaii followed by five in Africa, Asia, and Latin America.

Forty-five interns from 25 countries have spent from several weeks to six months at NifTAL acquiring skills while working in collaboration with NifTAL researchers. Additionally, NifTAL has either totally or partially supported 25 graduate students working toward advanced degrees.

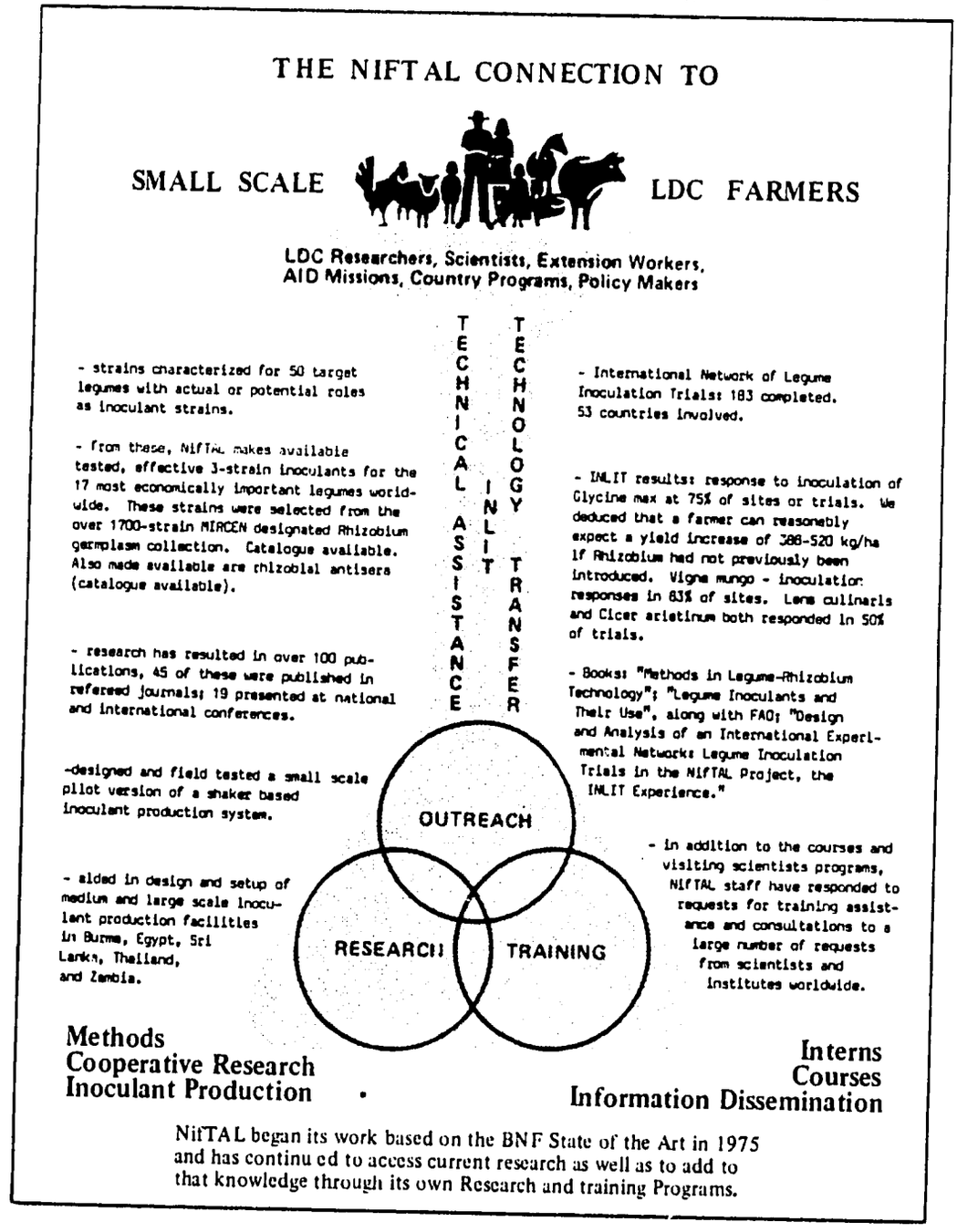
Two new training courses were instituted in 1984: BNF for LDC extension leaders (in conjunction with North Carolina State University and Bangladesh Agricultural Research Council) and an intensive Inoculant Production Course for LDC researchers and inoculant producers. NifTAL has also cooperatively sponsored several Azolla Technology Courses. Available in August 1985 will be the revised training manual **"Methods in Legume-Rhizobium Technology,"** the well tested result of NifTAL's training program.
This book will be on sale at the 10th North American Rhizobium Conference in August.

TRAINING RESULTS

This training has borne fruit as follow-up on ex-trainees has revealed that 60 are active in BNF research and outreach, 3 head BNF programs at National institutions, 13 are working toward advanced degrees, 10 are university professors, 2 are in policy-making positions, 18 have obtained outside funding to carry on BNF research, and 3 have offered their own BNF technology courses in cooperation with NifTAL.

The above presents just a few highlights of the many services that NifTAL offers. It has taken this integrated approach to keep abreast of changing needs and emphases in the development and promotion of BNF.

As NifTAL's tenth year passes, we can look back on many successes. We have learned, and through experience have adjusted our approaches, just as we have adapted to changing needs. Our momentum is up and a critical mass has been achieved. We are viable and fully ready to meet the challenge of our mission, to improve the lot of developing country farmers.



INDIVIDUALS FROM COUNTRIES OF THE NEAR-EAST AND ASIA WHO HAVE RECEIVED NifTAL TRAINING

A CUMULATIVE SUMMARY

Numbers of Participants by Country:

Bangladesh	11	Pakistan	3
Burma	10	Philippines	15
Egypt	5	Singapore	1
India	23	Sri Lanka	10
Indonesia	12	Sudan	4
Iraq	1	Taiwan	1
Malaysia	6	Thailand	38
Morocco	2	Tunisia	1
Nepal	1	Turkey	1

Number of Participants by Training Type:

Intern: 25
Short Term Intern: 17
Graduate Student: 4
Legume/*Rhizobium* Technology: 47
Legume Inoculant Production: 17
Azolla Technology: 20
Legume/*Rhizobium* Extension Services: 20

As of 1/24/86

INDIVIDUALS FROM COUNTRIES OF THE NEAR-EAST AND ASIA WHO HAVE RECEIVED NifTAL TRAINING

Key:
1 - Graduate Student
2 - Intern Program (Trainee & Visiting Scientist)
2A- Short Term Intern
3 - NifTAL/MIRCEN Nairobi Course (Kenya, 1979)
4 - Early Training Courses (NifTAL Maui, 1977 or 1978)
5 - NifTAL/MARDI Training Course (Malaysia, 1980)
6 - NifTAL Bangkok Training Course (Thailand, 1982)
7 - NifTAL/ICRISAT/FAO Inoculant Production Course (India, 1983)
8 - NifTAL Azolla Course (Thailand, 1984)
9 - NifTAL Inoculant Production Course (NifTAL Maui, 1984)
10 - NifTAL/NCSU Extension Course Prototype (Thailand, 1983)
11 - NifTAL BNFRC Inoculant Production Course (Thailand, 1986)

	NAME	CURRENT INSTITUTE	COUNTRY	TYPE	YEAR
1	Shamsul Hoque	Bangladesh Ag Univ	Bangladesh	4	1978
2	Mohd Shahjahan	BARI	Bangladesh	5	1981
3	Dilwara Begum	BARI	Bangladesh	6	1982
4	A.R.M. Solaiman	BINA	Bangladesh	6	1982
5	Sk. Md. Abdus Sattar	Dakar	Bangladesh	8	1983
6	Md Abdul Majid	Dakar	Bangladesh	8	1983
7	Md Idris Majlis	BARI	Bangladesh	2A	1984
8	Faisal A. Chowdhury	Bang. Agric Univ	Bangladesh	2A	1984
9	Muhammad Ali	B. Inst Nuclear Agric	Bangladesh	2A	1984
10	Shahidul Islam	BARI	Bangladesh	2A	1984
11	Taufiqul Aziz	Univ Hawaii	Bangladesh	1	Current
12	Daw Nwe Nwe Aung	Ag Res Inst Yezin	Burma	6	1982
13	Daw Khin San Wai	"	Burma	6	1982
14	U. Hla Than	"	Burma	2	1983
15	U Aung Khin	"	Burma	2A	1984
16	U Win Maung	Ag Corporation	Burma	2A	1984
17	U Kin Zaw	Ag Res Inst Yezin	Burma	2A	1984
18	Trillion Hmun	Ag Res Inst Gyogan	Burma	2A	1984
19	Tun Thein	Ag Corp (Moulmein)	Burma	2A	1984
20	Nuynt Wai	Ag Corp (Hmawki)	Burma	2A	1984
21	Shirley Smellie	Ag Res Inst Yezin	Burma	2A	1984
22	M. Essam Abd Elsamee	Plant Protection Dept Riyadh, Saudi Arabia	Egypt	4	1978
23	Mhd Ahmd Abd El Daiem	Agric Res Center (Giza)	Egypt	3	1979
24	Nabil A. Hegazi	King Saud U (Saudi Arabia)	Egypt	3	1979
25	Hussein M Abd Kamel	National Res Inst	Egypt	2	1982
26	Farouk M. Hammouda	Res Ext Ctr Kafr El Sheikh	Egypt	7	1983
27	B. Baskar Reddy	NCSU	India	2	1979
28	Neeru Narula	Haryana Agric Univ	India	4	1978

	NAME	CURRENT INSTITUTE	COUNTRY	TYPE	YEAR
29	N. V. Reddy	ICAR	India	4	1978
30	T. S. Sandhu	Punjab Agric Univ	India	2	1980
31	Ranbir Rewari	IARI	India	2	1981
32	S. R. Murthy	A.P. Agric University	India	5	1981
33	V. Reddy	ICAR Drlnd Proj Hyderabad	India	5	1981
34	Mayank Vora	Gujarat Ag Univ	India	2	1982
35	M. Rangarajan	Ctr Adv Studies Ag Micro	India	2	1983
36	G. R. Gajendragadkar	JNKVV	India	7	1983
37	F.Chand Garg	Panjab Univ	India	7	1983
38	Krishna Gupta	Haryana Agric Univ	India	7	1983
39	P. Mohan Khosla	Bacfil Inoculants	India	7	1983
40	Dulare Lal	Soil Testing Lab (Aligang)	India	7	1983
41	R. D. Mehta	Gujarat Fert Co, Ltd.	India	7	1983
42	B. A. Patel	Gujarat Co-op, Ahmenabad	India	7	1983
43	V. Basavaraju	Karnataka Dept of Agric	India	7	1983
44	S. K. Roy	State Ag Res Inst Calcutta	India	7	1983
45	S. Kannaiyan	Coimbatore	India	8	1983
46	N. R. Vishwanath	Bangalore	India	8	1983
47	K. S. Jauhri	IARI	India	2	1984
48	S. R. Hegde	Univ Ag Sci - GKVK Campus	India	4	1984
	Plus			2	1984
49	Thomas George	Univ Hawaii	India	1	Current
50	Gin A. W. Prauto	Bogor Agric Univ	Indonesia	4	1978
51	Edi Santosa	Ctr Soil Res (Bogor)	Indonesia	4	1978
52	Omar Hidayat	Food Crops Inst (Sukamandi)	Indonesia	2	1981
53	Ismael B. Poerboyo	Control Res Inst for Ag	Indonesia	5	1981
54	Tini Prihartini	Pasca Sarjana Padjadaram U	Indonesia	5	1981
55	Dia Purwantari	Inst Animal Sci	Indonesia	6	1982
56	Mursidi Sediyarso	Ctr Soil Res (Bogor)	Indonesia	2	1982
57	Dedah H. Arief	Bandung	Indonesia	8	1983
58	Soetjipto Partohardjono	Bogor	Indonesia	8	1983
59	Ruchyat Damanhuri	Bogor	Indonesia	8	1983
60	Muhrizal Sarwani	BARIF	Indonesia	9	1984
61	Muhrizal Sarwani	BARIF	Indonesia	2	1985
62	Ashar Al-Nashi	*	Iraq	2	1979
63	Irene Tan Kit Ping	Univ Malay	Malaysia	4	1978
64	Abd Karim bin Abd Ghani	Univ Kebangsaan	Malaysia	5	1981
65	S. M. bin Mohd Hashim	MARDI	Malaysia	5	1981
66	Fuzoyah bin Ibrahim	Rubber Research Inst	Malaysia	5	1981
67	Tan Guan Kok	Rubber Research Inst	Malaysia	5	1981
68	Noorani Malik	RISDA Kuala Lumpur	Malaysia	5	1981
69	Albrecht Glatzle	*	Morocco	9	1984
70	Ali Hilali	Inst Agronomique (Rabat)	Morocco	2	1986
71	Shiva Chaudhary	*	Nepal	1	1983
72	Mhd Akmal Siddiqi	Cornell U (Grad Student)	Pakistan	6	1982
73	Sikander Ali	Gordon College	Pakistan	8	1983
74	Naseer Mirza		Pakistan	8	1984
		Univ Hawaii		1	Current

	NAME	CURRENT INSTITUTE	COUNTRY	TYPE	YEAR
75	Gayaga Ocampo	Bureau of Soils	Philippines	4	1978
76	Fe Terres	Univ Phil. at Los Banos	Philippines	4	1978
77	Angela Almendras	Univ Phil. at Los Banos	Philippines	4	1978
78	Nora T. Armones	Bureau of Plant Industry	Philippines	6	1982
79	Crisanto Lopez	Univ Philippines (Diliman)	Philippines	6	1982
80	Visitacion Z. Perdido	Bur of Plant Ind. Iligan	Philippines	6	1982
81	Erlinda Paterno	College of Agriculture	Philippines	2	1982
82	Antonio Leviste	Manilla	Philippines	8	1983
83	Juan C. Bunoan, Jr	*	Philippines	8	1983
84	Celso R. Bersabe	*	Philippines	8	1983
85	Charles de la Torre	*	Philippines	8	1983
86	Abelardo Sorilla	*	Philippines	8	1983
87	Romy Necesario	*	Philippines	8	1983
88	Corazon Espinas	*	Philippines	8	1983
89	Rolando O. Torres	IRRI	Philippines	2	1985
90	Gloria Lim	National University	Singapore	2	1982
91	S.K.D. Aloysius	Reg Agric Res Station	Sri Lanka	4	1978
92	S. Logendra	CARI	Sri Lanka	4	1978
93	Chandini Mudannayke	Soybean Research Station	Sri Lanka	2	1981
94	Ponnampalam Thirokkumaran	Soybean Research Station	Sri Lanka	2	1981
95	S. Sivasibramanian	IDA Dairy Dev Project	Sri Lanka	5	1981
96	Wasanthaa L. Ratnayake	Gannoruwa	Sri Lanka	6	1982
97	Nirmala Gunapala	Colombo	Sri Lanka	8	1983
98	S. A. Kulasooriya	Colombo	Sri Lanka	8	1983
99	S. W. Abeysekera	Colombo	Sri Lanka	8	1983
100	Lal Senayake	Penn State Univ	Sri Lanka	2A	1984
101	Sayeda A. Abu Nayib	University of Khartoum	Sudan	3	1979
102	H. AbdelMagid	University of Khartoum	Sudan	2	1983
103	A. Ali Mahdi	University of Khartoum	Sudan	7	1983
104	A.G.E. El Shafie	University of Khartoum	Sudan	7	1983
	Plus			9	1984
105	N. S. Talekar	AVRDC	Taiwan	4	1978
106	Somsak Kotepong	Department of Agriculture	Thailand	4	1978
107	P. Smitamana	Cheing Mai University	Thailand	4	1978
108	Pornpun Jonsuksuntigool	Royal Forest Dept	Thailand	2	1980
109	Jessada Kaewchote	MAE-SA Project	Thailand	2	1980
110	Chirasak Arunsri	Dept of Agriculture	Thailand	5	1981
111	Bietsurak Bhokasawat	Pak Chong Forage Crop Sta	Thailand	6	1982
112	Chutip Chanaseni	Dept of Ag Ext Bangkhapi	Thailand	6	1982
113	Arawan Chutsrirung	Cheing Mai University	Thailand	6	1982
114	Wichian Magtoon	Srinakharinwiot Univ	Thailand	6	1982
115	Suganya Soontaros	Chulalongkorn Univ	Thailand	6	1982
116	Banyong Toomsan	Khon Kaen Univ	Thailand	6	1982
117	Ampan Bhromsiri	Cheing Mai Univ	Thailand	2	1982
118	Wichien Yongmanitchai	Kasetsart Univ	Thailand	9	1984
119	Taweesakdi Sesaweech	Dept of Agric Extension	Thailand	2A	1984
120	Awata Dalodom	"	Thailand	2A	1984
121	Pramual Setarat	"	Thailand	2A	1984

	NAME	CURRENT INSTITUTE	COUNTRY	TYPE	YEAR
122	Somsakdi Surya	Dept of Agric Extension	Thailand	2A	1984
123	Chavalvat Chainuvati	"	Thailand	2A	1984
124	Thawat Watkaew	North East Ag Ext Office	Thailand	10	1983
125	Anan Suriyegarn	Cent Reg Ag Ext Office	Thailand	10	1983
126	Pattamarat Redkachane	Dept of Ag Extension	Thailand	10	1983
127	Supote Chaivimol	Pichit Ag Office	Thailand	10	1983
128	Porntip Polapukde	Eastern Ag Ext, Rayong	Thailand	10	1983
129	Bancha Soparat	Seed Center 1, Pisanuloke	Thailand	10	1983
130	Prasat Stonsaovapak	Seed Center 2, Korat	Thailand	10	1983
131	Sompong Meeklai	Seed Center 4, Chainart	Thailand	10	1983
132	Narisara Kamragai	Seed Center 5, Lopburi	Thailand	10	1983
133	Pornchai Suthatorn	TKR Project	Thailand	10	1983
134	Anan Pintarak	Thai-Aust & World Bank Land Dev Project, Lampang	Thailand	10	1983
135	Chiraphant Laongsri	Nothern Reg Ag Dev Ctr	Thailand	10	1983
136	Vinita Atipirin	Southern Reg Ag Dev Ctr	Thailand	10	1983
137	Wirote Cholwiriyakul	Central Reg Ag Office	Thailand	10	1983
138	Waravuth Iampunpang	North East Reg Ag Dev Ctr	Thailand	10	1983
139	Sanayh Kraokau	Field Crops Res Inst	Thailand	10	1983
140	Preecha Prammance	Field Crops Res Inst	Thailand	10	1983
141	Somsak Srisombun	Srisumrong Fld Crp Exp St	Thailand	10	1983
142	Vudhisak Pornprompratan	Chieng Mai Fld Crp Exp St	Thailand	10	1983
143	Mujanart Kungpisadan	RRI, Hatyai	Thailand	10	1983
144	Mongi Zouaghi	Nat Inst Agric	Tunisia	9	1984
145	Velittin Gurgun	University of Ankara	Turkey	4	1978
	Plus			2	1985
	PENDING				
	M. Usha Kiran		India	11	1986
	Ujang Suparman		Indonesia	11	1986
	Usman Doras		Indonesia	11	1986
	Abdul Hamid		Indonesia	11	1986

* Current Address Unknown

INDIVIDUALS FROM COUNTRIES OF LATIN AMERICA WHO HAVE RECEIVED NifTAL TRAINING

A CUMULATIVE SUMMARY

Numbers of Participants by Country:

Argentina	3	Honduras	2
Bolivia	2	Mexico	13
Brazil	11	Nicaragua	1
Chile	1	Panama	4
Colombia	8	Peru	11
Costa Rica	11	Puerto Rico	2
Republic Dominicana	6	Trinidad	2
Ecuador	3	Uruguay	3
Guatemala	4	Venezuela	1

Number of Participants by Training Type:

Intern: 7
Graduate Student: 1
Legume/*Rhizobium* Technology: 64
Legume Inoculant Production: 2
Azolla Technology: 14

As of 1/24/86

INDIVIDUALS FROM COUNTRIES OF LATIN AMERICA
WHO HAVE RECEIVED NifTAL TRAINING

Key:
1 - Graduate Student
2 - Intern Program (Trainee & Visiting Scientist)
2A- Short Term Intern
3 - NifTAL/Colegio de Postgraduados Rhizobium Technology Course (Chapingo, Mexico, 1981)
4 - Early Training Courses (NifTAL Maui, 1977 or 1978)
5 - NifTAL/Porto Alegre MIRCEN Rhizobium Technology Course (Brazil, 1980)
6 - NifTAL/Porto Alegre Mircen/CIAT/UNDP/FAO Legume Rhizobium Systems (Brazil, 1985)
7 - NifTAL/CIPA II Azolla Course (Peru, 1983)
8 - NifTAL Inoculant Production Course (NifTAL Maui, 1984)
9 - NifTAL/Univ Costa Rica Rhizobium Technology Course (Led by former Intern, Costa Rica, 1983)

	NAME	CURRENT INSTITUTE	COUNTRY	TYPE	YEAR
1	Alejandra T. Ayala Torres	Univ Buenos Aires	Argentina	5	1980
2	Luisa del Valle Oliva	Ciudad Univ	Argentina	5	1980
3	Ricardo O. Russo	Univ Buenos Aires	Argentina	9	1983
4	Eduardo Borenstein	CORGEPAI	Bolivia	9	1983
5	Renato V. Bedregal	CIAT	Bolivia	6	1985
6	Edemar Brose	EMPASC-ESTACAO	Brazil	2	1980
7	Marcio Voss	IAPAR	Brazil	5	1980
8	Pedro A. Arraes Pereira	CNPAF, Goiania	Brazil	5	1980
9	Roberto Bonetti	CENA/UPS	Brazil	5	1980
10	Luis Antonio de Oliveira	Inst Nat Pesquisas Amazon	Brazil	5	1980
11	Marcelo G. de Moraes	IPAGRO	Brazil	9	1983
12	Nuno M. de Souza Costa	EMBRAPA	Brazil	2	1979
13	Marli de Fatima Fiore	EMBRAPA	Brazil	7	1983
14	Catharina Chou Pan	Agro-Quimica Plto, Ltda	Brazil	8	1984
15	Stela Vieira Midlej Silva	CEPLAC/CEPEC/DIFIT	Brazil	6	1985
16	Reinaldo B. Cantarutti	CEPEC	Brazil	6	1985
17	Emma M. H. T. Montalbetti	INTEC - Chile	Chile	5	1980
18	Raul Varella Gonzalez	ICA/AA	Colombia	5	1980
19	Yolanda N. de Navarro	Ciudad Univ	Colombia	5	1980
20	Ernesto Luque	Univ de Narino	Colombia	9	1983
21	Hector Laverde	ICA/La Libertad	Colombia	7	1983
22	Edmundo Garcia	ICA/Palmira	Colombia	7	1983
23	Fernando Munevar	ICA/Tibaitata	Colombia	7	1983
24	Joaquin G. Pena	ICA, Codazzi	Colombia	6	1985
25	Justo A. Barros Henriquez	ICA/CIR, Motilonia	Colombia	6	1985
26	Oscar Acuna Navarro	Univ de Costa Rica	Costa Rica	3	1981

	NAME	INSTITUTE	COUNTRY	TYPE	YEAR
27	Maria del Rocio Lopez V.	Escuela de Biologia	Costa Rica	9	1983
28	Eduardo Mata Montero	CARE	Costa Rica	9	1983
29	Benjamin Mora G.	Inst Tech de Costa Rica	Costa Rica	9	1983
30	Oscar J. Rocha Nunez	Escuela de Bologia	Costa Rica	9	1983
31	Patricio Solis Barrantes	Center of Invest Agron	Costa Rica	9	1983
32	Leonidas Villalobos	Sch of Agric Science	Costa Rica	9	1983
33	Carlos Ramirez	Centro Invest. Agronom.	Costa Rica	2	1981
34	German H. Cordoba	CNP, Sabana	Costa Rica	5	1985
35	Lidieth Uribe Lorio	University of Costa Rica	Costa Rica	6	1985
36	Jose Murillo Vargas	Ministry of Agricultue	Costa Rica	7	1983
37	Mercedes Perez G	Univ Autonoma S. Domingo	R. Dominican	5	1980
38	Hector Rafael Reynoso	Univ Autonoma S. Domingo	R. Dominican	3	1981
39	Manual Rodriquez	Inst Sup Agricultura	R. Dominican	7	1983
40	Gustavo Pena	CEDIA/Juma	R. Dominican	7	1983
41	Carlos G. Suarez	INDOTEC	R. Dominican	8	1984
42	Joaquin M. Guerrero	Dept of Agriculture	R. Dominican	9	1983
43	Jose Reinaldo Unda Calarza	INIAP	Ecuador	5	1980
44	Carlos H. P. Jaramillo	Ministry of Agriculture	Ecuador	3	1981
45	Flavio G. P. Badillo	Exp Station Sta Catalina	Ecuador	9	1983
46	Rolando G. Aguilera Majia	Ciudad Univ	Guatemala	5	1980
47	Hugo Abel de la Cruz D.	Univ San Carlos	Guatemala	3	1981
48	Marcial E. G. Alburez	ICTA	Guatemala	6	1985
49	Maria Ines Castellanos A.	ICAITI	Guatemala	6	1985
50	Ana Margoth Chavez A.	Esc Agricola Panamerica	Honduras	3	1981
51	Reina G. Flores Nunez	Ministry of Nat Resources	Honduras	3	1981
52	Ronald Ferrera-Cerrato	Univ of Puerto Rico	Puerto Rico	4	1978
53	Gaspar Reyes Gonzales	Inst Nat invest Pecuarias	Mexico	5	1980
54	Abdul Khalil Gardezi	Cent. de Edafologia	Mexico	9	1983
55	Jorge Peisajovich	Univ of Mexico	Mexico	9	1983
56	Alejandro P. Vasquez	Univ of Puerto Rico	Mexico	9	1983
57	Eduardo L Alcocer	Postgraduate College	Mexico	3	1981
58	Ana L Sanchez A.	Bank of Mexico	Mexico	3	1981
59	Adolfo D A Bojorguez	Univ de Sinalra	Mexico	3	1981
60	Maria G Tsuzuki Reyes	Nat Univ Autonoma	Mexico	3	1981
61	Arturo C. Espinoza	Quimica Lucava	Mexico	9	1984
62	E. Cuautle-Fabian	Postgraduate College	Mexico	1	1983
63	Joel Espinosa Monteros	SEP Inst Technologico	Mexico	2	1982
64	Juan F. Aguirre Medina	INIFAP	Mexico	6	1985
65	Anselmo C de Aquino	INIFAP	Mexico	6	1985
66	Francisco Ruiz	Dir Nat Tech Agriculture	Nicaragua	3	1981
67	Blanca Hernandez	Univ of Panama	Panama	4	1978
68	Humerto J. Cornejo	Univ of Panama	Panama	3	1981
69	Estegan A. Jaen	IDIAP	Panama	6	1985
70	Margarita X C Montenegro	Univ of Panama	Panama	6	1985
71	Asuncion Cano Echevarria	Univ Nac Mayor San Marcos	Peru	5	1980
72	Judith I. la Rosa la Rosa	Univ Nat Cajamarca	Peru	3	1981
73	Tomas Tello Reyes	Exp Sta/Vista Florida	Peru	2	1981
74	Luis Arevalo	Univ National	Peru	7	1983

	NAME	INSTITUTE	COUNTRY	TYPE	YEAR
75	Angel Diaz Celas	Univ National	Peru	7	1983
76	Jose Lopez	Exp Sta/San Ramon	Peru	7	1983
77	Frederico Minguillo	CIPA II	Peru	7	1983
78	Andres Serquen	CIPA X	Peru	7	1983
79	Guillermo Prado Beneju	Nat Univ San Agustin	Peru	9	1983
80	Silos G. del Aguila	IVITA	Peru	6	1985
81	Cesar A. Reys Atac	IVITA	Peru	6	1985
82	Yovanny Ant. Valazquez	Univ of Puerto Rico	Puerto Rico	9	1983
83	Alfred Donawa	Univ West Indies	Trinidad	2	1980
84	Richard Graham	Univ West Indies	Trinidad	4	1978
85	Amalia Lucas de Villamil	Plan Agropecuaria	Uruguay	5	1980
86	Enriquez Deambrosi	CIA/Alberto Boerger	Uruguay	7	1983
87	Carlos Labandera	Plan Agropecurio (MAP)	Uruguay	7	1983
88	Vicente A. Michelena	Univ de Oriente	Venezuela	2	1982

INDIVIDUALS FROM COUNTRIES OF AFRICA WHO HAVE RECEIVED NifTAL TRAINING

A CUMULATIVE SUMMARY

Number of Participants by Country:

Benin	1	Mauritania	1
Ethiopia	1	Nigeria	6
Ghana	4	Rwanda	1
Guinea Bissau	1	Senegal	3
Ivory Coast	1	Sierra Leone	2
Kenya	5	Tanzania	4
Liberia	2	Upper Volta	1
Mali	3	Zambia	2

Number of Participants by Training Type:

Intern: 9
Graduate Student: 3
Legume/Rhizobium Technology: 15
Legume Inoculant Production: 1
Azolla Technology: 11

As of 1/24/86

INDIVIDUALS FROM THE COUNTRIES OF AFRICA
WHO HAVE RECEIVED NifTAL TRAINING

Key: 1 - Graduate Student
2 - Intern Program (Trainee & Visiting Scientist)
2A- Short Term Intern
3 - NifTAL/MIRCEN Rhizobium Technology Course (Kenya, 1979)
4 - Early Training Courses (NifTAL Maui, 1977 or 1978)
5 - NifTAL Azolla Course (Senegal, 1984)
6 - NifTAL BNFRC Inoculant Production Course (Thailand, 1986)
NOTE: Other Courses have been mounted at the University of Nairobi, Kenya, by participants--led by Dr. S. O. Keya--who participated in Course number 3 above.

	NAME	CURRENT INSTITUTE	COUNTRY	TYPE	YEAR
1	Moustapha Adomou	*	Benin	5	1983
2	Michael Akyeampong	*	Ghana	1	1980
3	Amare Abebe	Ag Res Inst/Nazareth	Ethiopia	2	1982
4	James Quashie-Sam	Univ of Ghana	Ghana	4	1977
5	M. S. Elegba	Univ of Ghana	Ghana	3	1979
6	Felix D. Dakora	Crops Res Inst/Nyankpala	Ghana	5	1983
7	Frank Sorwli	Univ of Ghana	Ghana	2	1984
8	Alberto Sanca	*	Guinea Bissau	5	1983
9	Wole Simeon Zirumba	*	Ivory Coast	5	1983
10	Chege Mugane	Univ of Nairobi	Kenya	4	1977
11	G. R. Mathenge	Plant Breeding Sta/Njoro	Kenya	3	1979
12	S. Wangarıro	Univ of Nairobi	Kenya	3	1979
13	F. Njeru	Univ of Nairobi	Kenya	3	1979
14	James N. Chui	Kenya Ag Res Inst	Kenya	2	1984
15	Wilson Kagobo	Univ of Hawaii	Liberia	1	Current
16	Alonzo N. Munyeneh	*	Liberia	5	1983
17	Kalilou Diakite	*	Mali	5	1983
18	Abdoulaye Traore	*	Mali	5	1983
19	Seydou Sanogho	deceased	Mali	2	1983
20	Dramane Kamara	*	Mauritania	5	1983
21	Lawrence Osa-Afiana	Ahmadu Bello Univ	Nigeria	4	1977
22	W. A. Osinaike	Moor Plantation	Nigeria	4	1978
23	John O. Ekpe	Univ of Ibadan	Nigeria	3	1979
24	Jolade Ayodele	*	Nigeria	5	1983
25	Sheriff O. Sanni	*	Nigeria	5	1983
26	Amos Ajayi	Univ of Ibadan	Nigeria	2	1982
27	A. Hakizimana	I.S.A.R. Rubona	Rwanda	3	1979
28	H. F. Diara	*	Senegal	5	1983
29	Mateugue Diack	*	Senegal	5	1983

	NAME	INSTITUTE	COUNTRY	TYPE	YEAR
30	Birane Kane	*	Senegal	5	1983
31	Charles H. Dixon	*	Sierra Leone	5	1983
32	Dennis Amara	Njala Univeristy	Sierra Leone	2	1981
33	Manase Salema	Univ Dar Es Salaam	Tanzania	4	1977
34	Ernest Semu	Univ Dar Es Salaam	Tanzania	4	1978
35	G. P. Msumali	Univ Dar Es Salaam	Tanzania	3	1979
36	Anselm Lwoga	Univ of Dar Es Salaam	Tanzania	2	1980
37	Komi Senyenarro Hovi	*	Togo	5	1983
38	Alfred Traore	*	Upper Volta	3	1979
39	Martin N. Nbewe	Ministry of Agriculture	Zambia	6	1985
40	R. C. Nyemba	Mount Makulu Cent Res	Zambia	3	1979
	Plus			1	Current
	Plus			6	1986

* Current Address Unknown

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IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX D - STATE-OF-ART

Revised:4/21/86: Revised 5/8/86
DIMPEX

• STATE OF THE ART

Introduction

Nitrogen is an essential constituent of proteins and large amounts of it are required by all forms of life. Plants provide protein to other forms of life, but for virtually all food crops, synthesis of their own protein depends on nitrogen compounds present in the soil. The amount and availability of the soil nitrogen to plants is conditioned by biological processes in the soil.

Nitrogen and other essential nutrients removed from the soil must be replenished, in one way or the other, to ensure sustained agricultural productivity. This can be accomplished through addition of chemical fertilizers which are costly and require heavy capital investment for production, storage, transportation and application. For example, the industrial synthesis of nitrogen requires large quantities of hydrogen gas from fossil fuel plus additional energy to maintain the high temperatures and pressures necessary to chemically combine nitrogen gas, N_2, with hydrogen, H_2, to yield ammonia, NH_3. In contrast to this, there is a diversity of biological organisms that can satisfy their nitrogen needs by directly incorporating into their proteins nitrogen gas (N_2) which is the major constituent of air; a process referred to as biological nitrogen fixation (BNF). Some of these organisms can live in close association with leguminous crop and tree species and contribute to their N economy. Soybeans, beans, and Leucaena are well-known examples of food and fuelwood products belonging to the legume family. Legumes are particularly attractive when sustained food and fuelwood productivity is sought from low-input farming systems. This is because of their high nutritional quality and their unusual ability to be self-sufficient for nitrogen supply.

The biological nitogen fixation (BNF) process

Biological nitrogen fixation (BNF) in legumes is possible by virtue of the mutually beneficial association (symbiosis) which can form between leguminous plants and certain microorganisms from a specific family of soil bacteria known as Rhizobium. Rhizobia can penetrate the roots of legumes and give rise to highly specialized organs referred to as root-nodules. These are quite different from tumors or other swellings that commonly occur on plant roots as a result of infection by disease-causing (pathogenic) organisms. The structure and function of nodulated legumes is modified in such a way that carbohydrates (sugars) produced in the leaves of the plant during photosynthesis are delivered to the nodulated root where they are respired to provide energy. In the nodules, this energy is consumed to maintain conditions under which nitrogen fixation can proceed and also to sustain the growth requirements of the rhizobia.

Gaseous dinitrogen enters the nodule from the air spaces in the surrounding soil. An enzyme, nitrogenase, which is the unique contribution of the microsymbiont, catalyses the splitting of dinitrogen molecules and the reaction of their component atoms to form ammonia. Neither the sequence of

reactions and transformations that follow initial fixation nor the precise sites in the nodule where the events occur are fully understood. The steps involve very rapid incorporation of ammonia, which would ordinarily be toxic to both symbionts, into nitrogenous compounds such as amino acids, amides and/or ureides depending on the particular legume species. These are removed from the nodule in the plant's transpiration stream to be used throughout the plant as building blocks for plant proteins.

Agrotechnologies based on BNF by legumes

Most farmers in the tropics do not know that legumes fix nitrogen in their root nodules. Yet traditional and modern farming systems of the tropics almost invariably include legumes (1,2). Thus legume cultivation results from recognition by farmers over many centuries that legumes are valuable components in farming systems rather than from intentional exploitation of biological nitrogen fixation *per se*.

Agrotechnology based on BNF by legumes has therefore two major aspects. One is related to the deliberate inclusion of legumes in cropping systems to derive benefits from their nitrogen fixation. The other concerns the intentional use of specific practices to maximize nitrogen fixation by legumes. For convenience these two facets of BNF technology will be referred to as "use of legumes" and "inoculation technology". The distinction is drawn to emphasize that currently legumes are utilized widely with less than maximal benefits from BNF because of deficient symbiotic associations. Productivity could be increased through use of appropriate technology to assure effective symbiotic nitrogen fixation by legumes in the role they currently play in the agricultural sector. Much greater gains in productivity and economies of energy from reduced fertilizer requirements will be realized through innovative use of legumes in roles they have not occupied previously in production systems, e.g., the use of fast-growing leguminous trees in agroforestry systems. Production gains will be greatest if the use of legumes is always complemented by appropriate inoculant technology. This is because legumes can only benefit fully from biological nitrogen fixation if they encounter rhizobia with which they are genetically compatible, so that root infection and nodulation can occur and if the strain which forms nodules functions effectively in fixing nitrogen.

The use of legumes

The benefits from BNF through the use of legumes in farming systems are both direct, because the legume has an intrinsic value, and indirect, as inclusion of a legume affords greater yield stability in adverse growth conditions and can benefit companion or following non-leguminous crops. Direct benefits from biological nitrogen fixation by legumes in cropping systems arise from the multiple uses of plants in the legume family. Though known primarily for grain, forage or feed production, legumes are also cultivated in the tropics for timber, fuelwood, green manures, oils, fibers, gums, drugs, dyes, and resins. Additionally, they may be used as hedges, ground covers for weed, insect and disease control, as soil stabilizers on terraced slopes or simply for shade or as ornamentals (3).

Indirect benefits accrue from the stability of performance and assurance of some economic return for at least one component under unfavorable conditions when legumes are intercropped with other crops. Stability is afforded, for example, in erratic rainfall zones when the components in the intercropping system are separated in time such as with sorghum/pigeon pea and groundnut/cotton (4,5). When there is an outbreak of pests or diseases, maize/beans and other intercrops afford stability of yields and income (6,7). Other indirect benefits accrue from the ability of legumes to make a net contribution of nitrogen to the soil under some circumstances, thereby reducing the N-fertilizer requirement for a companion or following non-leguminous crop.

Inoculant biotechnology

There is a commonly held view (8) that tropical legumes are much more promiscuous than temperate legumes in that they nodulate freely with a wide range of tropical rhizobia; and that tropical soils are laden with such bacteria to an extent which virtually guarantees effective nodulation without inoculation (9,10). This view is no longer well-founded. Some species and accessions from genera previously considered to be promiscuous (8) require specific strains of *Rhizobium* (11,12,13) or form highly effective symbioses with only a few out of the wide array of strains with which they nodulate (14,15,16). Recent intensification of interest in the tropical legumes and their rhizobia is revealing much greater variation in genetic compatibility and nitrogen fixation effectiveness among tropical rhizobia than has generally been acknowledged (17,18). A plan has been made for recognition that tropical legumes fall into one of three categories (18):

Promiscuous effective (PE) group in which nodulation occurs with a wide array of rhizobia isolated from many legume genera and the resultant symbioses are predominantly effective in nitrogen fixation.

Promiscuous ineffective (PI) group in which nodulation occurs with an array of strains of rhizobia isolated from many legume genera, but in which fully effective symbioses form with only a few of those strains.

Specific (S) group in which only those strains from the same genus (or, commonly, a restricted number of other genera) form effective symbioses.

Just as with the temperate legumes, the likelihood that compatible effective rhizobia will not always be encountered already present in sufficient numbers in the soil microflora is the basis for inoculation technology for tropical legumes (19). When a tropical legume seed is sown uninoculated in a tropical soil, a native rhizobial population of strains differing greatly in their symbiotic effectiveness compete for the finite number of nodulation sites on the legume roots. Many forage legumes bear only 10-20 nodules on which they depend for nitrogen during the first three months of their establishment. Thus it becomes critically important that each of the nodules which form on the root contain a strain of Rhizobium that is fully effective in fixing nitrogen. The underlying objective in inoculation technology is to introduce sufficiently high numbers of preselected strains of rhizobia into the vicinity of the emerging root that they have a competitive advantage over any indigenous soil strains of lesser N-fixing ability in the formation of root-nodules.

Inoculation technology involves: selection of strains of rhizobia that are compatible and effective N-fixers with particular legumes; multiplying selected strains to high population densities in bulk cultures; incorporating the liquid rhizobial cultures into a carrier material (usually finely milled peat) for packaging and distribution; and finally, coating the seeds of legumes with the carrier or implanting the soil with the inoculant directly into the seed drill (16,20,21).

An inoculum strain of Rhizobium recommended for a particular host must be able to form effective N-fixing nodules with that host under a wide range of field conditions. Nitrogen fixation effectiveness is only one important criterion for an inoculant strain. Other criteria include: competitiveness in nodule formation, particularly against less effective strains; persistence in the soil in the absence of the host, especially for strains for annual species; promptness to form nodules; ability to fix nitrogen under a range of soil temperature conditions; tolerance to pesticides; tolerance of low soil pH; nodulation in the presence of high levels of soil nitrogen; and ability to grow and survive in the peat inoculants.

The host genotype interacts with the infecting strain of Rhizobium in determining the level of nitrogen fixation with the host playing the dominant role. Thus two sources of variation (plant and Rhizobium strain) can be exploited in selection programs. Most commonly, though, the plant is selected independently and a suitable strain sought thereafter, thus allowing only for exploitation of strain variability. The range of specificities of host genotype interactions is well illustrated by soybean (22) and the African clovers (23).

Such specificities give three options in the approach to selection of strains for inoculants: numerous inoculants, each with a highly effective strain for inividual species; 'wide-spectrum' strains that vary from good to excellent in N_2-fixation with a range of legumes; or multiple-strain inoculants containing the best strain for each host species. There may be a conflict between the option that would be chosen for commercial expediency and that which is scientifically excellent (24). In Australia 'wide-spectrum' strains are used when these are available, but there is increasing use of specialized inoculants with specific strains for

individual hosts. Despite findings which suggest that multi-strain inoculant should be avoided because of possible antagonistic and competitive effects in culture (25) and the likelihood of competition in nodule formation from the less effective strains (26), this is the approach used successfully by the U.S. inoculant industry.

Strains for testing can be obtained from other laboratories working with the same species, from nodules on plants in the native habitat from which they were originally collected for plant-introduction/evaluation programs, and from nodules formed on the legume by native strains after sowing uninoculated seed in the region where the new species is expected to be used. None of these sources is invariably better than the other in screening programs.

Most legume inoculants are prepared by adding liquid cultures of Rhizobium to a finely-ground carrier base material such as peat. Although mixtures of peat with soil or compost mixtures, lignite, coir dust and some other organic materials have been used, peat has proven to be the most acceptable carrier worldwide. Agar, broth and lyophylized cultures are not recommended because of the very poor survival of these forms of the inoculum on seed (27,28,29).

Peat cultures can be prepared in two ways. Either ground (milled) peat is mixed with a high viable count (more than 10^9 rhizobia/ml) broth culture in sufficient volume to provide the minimum number of Rhizobium acceptable for use, or sterilized peat is inoculated with a small volume of culture and incubated to allow multiplication of the rhizobia in the carrier. The choice of method will depend on two main factors -- the survival of the rhizobia in peat in numbers high enough to meet a minimum standard of quality, and the availability of suitable, sterilizable containers and sterilizing facilities. The two factors that most affect survival of rhizobia in peat are temperature of storage and sterility of the peat. There are differences among species and also between strains of the same species of Rhizobium in their ability to survive well in peat (30).

Like all biological products, legume inoculants are prone to loss of quality owing to variation in the organism concerned and from unforeseen factors affecting some aspect of growth or survival. It is therefore essential that a quality control system be established. In Australia large-scale manufacture of legume inoculants is by private enterprise, and a separate, official (government) control laboratory is responsible for maintaining a high quality product. The control laboratory maintains and supplies recommended strains of Rhizobium to the industry, checks strains annually for ability to fix N_2, assesses quality of cultures during and after manufacture, and conducts such research as may be necessary to overcome problems associated with the production and survival in the final product. In the U.S. the industry is free to select its own strains and officials control ensures that the product can form nodules on the legume for which it is recommended.

Although control of quality of inoculants is primarily in the manufacturer's interest and therefore his responsibility, power of control by external bodies provides protection from less scrupulous operators and

genuine failure of a strain outside manufacturer control. Not all countries back their control labs with legislation. A control requires suitably qualified and experienced personnel with facilities to permit normal aseptic culture transfer and plant growth facilities suitable for legumes over the range for which inoculants are manufatured. Methods of assessments involve both qualitative and quantitative tests. The number and extent of these may vary according to competence and experience of manufacturers and of the standards desired. In Australia this control extends to holding stocks of the strains used in inoculants. This is not the case in the U.S. (13,31). In addition to assessment of quality throughout manufacture, it is important to monitor quality of product in retail outlets. Standards acceptable at this level may vary from that at manufacture and between countries. It is important that standards be realistic and within the capability of manufacturers yet ensure that sufficient viable rhizobia are applied to the seed to provide a satisfactory inoculation. In many instances this can be as few as 100 rhizobia per seed but in case of severe environmental stress as high as 10,000 or even 500,000 (28,32,33).

The prime objective of inoculation of legume seed with rhizobial inoculants is nodulation of the introduced legume host plant. Rhizobia introduced into new environments must live saprophytically in competition with other rhizobia and soil microorganisms in an environment which may be adverse for their growth and survival, until such time as the host seedling roots provide the ecological niche to which they are adapted. Thus steps should be taken to ensure that inoculant strains: remain viable until the host seedling is at the susceptible stage for infection; compete with any naturalized rhizobia for infection sites on the roots of the host legume and so form sufficient nodule tissue to permit maximum nitrogen fixation; nodulate its host promptly and effectively over a range of environmental conditions; and persist in the soil for at least several years in sufficient numbers to maintain nodulation of perennial legumes or to achieve prompt nodulation of regenerating annual species.

The first attempts at inoculation involved the transfer of soil from one field to the next, but with the isolation of the organisms responsible for nodule formation, artificial cultures soon replaced the laborious soil transfer technique. The usual inoculation technique is to apply the inoculant to the seed just before sowing either as a dust or as a slurry with water or adhesive solution. Adhesives such as gum arabic and substituted celluloses not only ensure that all the inoculum adheres to the seed surface but also provides a more favorable environment for survival of the inoculum. Pelleting of seed with finely ground coating materials such as lime, bentoninte, rock phosphate and even bauxite (32,34) have been used to protect rhizobia during their time on the seed coat. Pelleting is a simple on-farm technique (32,35) but custom-pelleted (by seedsmen at farmer's request) and preinoculated seed is now more popular. This latter procedure is potentially able to provide high populations of rhizobia on the seed for long periods of time (one growing season to the next) but has not yet been fully developed or exploited. Most preinoculation procedures are based on multiple coatings, alternately of adhesive and finely ground pelleting materials as used in simple pelleting. The peat inoculant is included as one (or more) of these coating layers. Soaking seeds in a

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broth suspension and then exposing them to either high pressure or vacuum to impregnate the rhizobia into or below the seed coat has not proven successful. Theoretically, rhizobia introduced in this way would be protected from drying and other adverse environmental conditions, but the quality of products produced commercially has been variable to very poor (36,37,38). It is, in fact, an indictment of the research workers in this area that 25 years has yielded so little progress in an area that has so much to offer for those concerned with the practical aspects of agricultural microbiology. The technique is particularly applicable to the less well developed and inexperienced rural groups in that a high quality and reliable product could be marketed by a manufacturer or distributor thus eliminating the need for farmer involvement in legume inoculation.

An alternative to pelleting and preinoculation in recent years has been the use of concentrated liquid or solid granular peat culture which can be sprayed or drilled directly into the soil with the seed during planting. Suspensions of rhizobia either as reconstituted frozen concentrates or suspensions of peat inoculant can be applied with conventional equipment. Similarly, granulated peat inoculants can be drilled in from separate hoppers on the drilling equipment. These methods have been especially successful for introducing inoculant strains into situations where there are large populations of competing naturally occurring soil rhizobia (39) or in cases of adverse conditions such as hot-dry soils (40) and where insecticide or fungicide seed treatment precludes direct seed inoculation (33,41). Solid inoculant, also known as granular or "soil implant" inoculum, is advantageous also, where seeding rates for crop legumes of 70-100 kg/ha make on-the-farm inoculation logistically impracticable.

Current use of legume-based BNF technology

(a) The use of legumes: The grain legumes are cultivated widely in a variety of agro-climatic zones in the tropics and subtropics. Total area in grain legumes in 1979 was 175 billion hectares. Dry bean (*Phaseolus vulgaris*) is the most important grain legume in Latin America, groundnut (*Arachis hypogaea*) in Africa and collectively groundnut, pigeon pea (*Cajanus cajan*)and chickpea (*Cicer arietinum*)in Asia. These and other grain legumes have been consistent components of human diet in the tropics for centuries yet in quantitative terms they continue to be minor crops.

The use of legumes in mixed legume/grass pastures in the tropics is at present restricted to northern Australia, the United States (Hawaii, Florida), southern Brazil and northeastern Argentina. The total area in improved legume/grass pasture is insignificant compared to the area of native grasslands under grazing. The use of temperate forage legumes in mixed pastures at high altitude locations in developing countries within the tropics is feasible but is considered outside the scope of this report.

Production statistics for the tropical grain legumes are seldom accurate. Most of the production is on a subsistence scale on small farms and the yields are seldom included in official statistics. Thus a figure of 186 million tons (42) should be regarded as an understatement.

There are very many agencies supporting and conducting research related to the use of legumes. International agencies such as FAO, UNDP, IBPGR, and the IARCs all have grain and forage legume programs. The USAID together with the govenmental agencies of many countries engaged in foreign agricultural development support research on legumes. The World Bank and several private and public foundations also support legume research. The author is not aware of any country in the tropics that does not have a legume project within its official agricultural program. Additionally, universities and agricultural colleges in tropical countries usually have legume programs. These projects cover the physiology, plant nutrition, agronomy, pathology, entomology, breeding and seed production of legume crops. Insofar as BNF proceeds at a rate governed strongly by the plant's ability to deliver carbohydrate to its root nodules, most technologies that improve overall plant performance are likely to have a beneficial impact on nodulation and nitrogen fixation by the respective legume. In fact some research programs with legumes are conducted under nitrogen-fertilized conditions or in fertile, N-rich soils. Breeding for high-yielding varieties under such conditions has resulted in plant types which are only weakly symbiotic and heavily dependent on soil nitrogen; e.g., bush bean (*Phaseolus vulgaris*) varieties developed in the United States.

Given the important role of grain legumes as the major dietary protein source for low-income groups in the developing countries, it is hardly surprising that such a multitude of funding agencies and implementing organizations give attention to research on legume technology. While it is to be expected that there will be overall gains in the quantities of nitrogen fixed from improved performance by legumes in the roles, and on the acreage they currently occupy, the major gains in BNF through the use of legumes will follow increases in the total land area in which legumes are grown and especially the innovative use of hitherto under-utilized legumes.

(b) Inoculant technology: Inoculant technology is implemented widely on a commercial scale mainly in the developed countries, the United States and Australia having substantial industries for the production, distribution and marketing of legume inoculants. There is also commercial-scale production in Brazil, Uruguay, Argentina, India and Egypt. Inoculants are available commercially in many other countries but produced in U.S. or Australian laboratories for importation and marketing by a subsidiary of the parent company or suppliers of legume seed, e.g., the Nitragin Company, Milwaukee, has subsidiaries in Mexico, Venezuela and Argentina and distributes inoculants worldwide. Some research centers, such as CIAT and the University of Hawaii NifTAL Project produce inoculants in pilot-scale plants as a service mainly to researchers and occasionally to legume growers. Demands for inoculation technology are increasing, associated mainly with the increased use of soybeans. There are dangers in trying to satisfy this demand by importation of inoculants developed in the U.S. or elsewhere. This is because inoculation technology as presently praticed has not proven transferable. That is to say, strains of *Rhizobium* and inoculation methods developed for conditions at one location in a particular farming system do not perform equally well at another location in a different farming system. Furthermore, the viability of rhizobia in

legume inoculants is greatly affected by storage conditions during shipment. Since producers are unable to control such factors, no guarantee can be given that the inoculants are of merchantable quality on arrival at their destination. For both these reasons inoculation failures are a common occurrence and this is adversely affecting consumer acceptance of the technology. An ideal scenario for improved implementation of BNF technology is described in a later section.

Those organizations funding research to adapt inoculant technology to the circumstances in which it will be used in tropical countries are: UNDP by its support to the IARC's through CGIAR and for a specific research program involving IITA and BTI/Cornell University; UNEP and UNESCO support inoculant technology under MIRCEN Project; FAO is actively considering the role it might play in the adaptation of inoculant technology of use in developing country agriculture (consultancy reports have been prepared and a planning workshop held during 1980); USAID through contracts with University of Hawaii (NifTAL Project) and USDA, Beltsville ARC (World Rhizobium Study and Collection Center) through grants under Section 211-(d) to the U.S. Universities' Consortium on BNF in the tropics, and through a portfolio of small grants administered by USDA SEA/CR; USAID and several governmental and non-governmental agencies who support the CGIAR are thereby sponsoring work at CIAT, IITA, ICRISAT and ICARDA on the adaptation of inoculant technology for use in the tropics.

How BNF by legumes increases crop yields and soil fertility

Consideration of the possible pathways for transfer of N from legumes to other crops (Fig. 1) affords a model around which estimates of the relative importance of the pathways of transfer of N from legumes to other crops and/or the soil can be attempted. Nitrogen gains per hectare per year entering the cycle as seeds, 1-2 kg (43) and in acid rainfall, 1.5 - 3.5 kg (44) are small compared to the nitrogen accumulated from the soil fixed biologically. About 50% of the nitrogen accumulated in legumes in fertile soils is attributed to BNF(45) though the proportion from fixed nitrogen will be greater in impoverished soils and lesser under nitrogen fertilization. Nitrogen accumulation in legume monocrops ranges from 50 - 350 kg/ha/year. It is generally accepted that fixation of around 100 kg/ha can be expected from the majority of grain and forage legumes. High levels are typically those of leucaena and other forage legumes with a twelve-month growing season. Low levels are typically those of notoriously bad nitrogen fixers with short growing seasons (e.g., Phaseolus vulgaris). As an illustration, the fate of, say, 100 kg of biologically fixed N entering the cycle will now be followed. Between 60 - 90% of the nitrogen accumulated in legumes is removed as grain depending on the species, harvest index and harvesting practice, or as animal products depending on the intensity and selectivity of grazing. Thus in an intercropping system only 10-40 kg nitrogen could potentially benefit other crops. Some of the organic nitrogen of the legume residues is mineralized rapidly and the rest is added to the soil organic matter pool from which it is mineralized slowly over a much longer period. In the studies that have been performed, 60% is probably the maximum portion of the nitrogen in the organic residue of a legume crop that could be mineralized in time to benefit a following crop.

'Using 50% initial mineralization for the purposes of these calculations, in a cropping system in which the legume fixes 100 kg/ha/year only 5 - 20 kg of nitrogen is likely to benefit the following crop. One practice that would substantially increase the contribution is green manuring. On the basis of the level of nitrogen fixation cited above (100 kg/ha/year), a residual benefit of 50 kg/ha/year to a following crop could be anticipated after incorporation of one year's production into the same area prior to raising the following crop. Greater benefits still can be contemplated if the green manure from a large production area is carried for incorporation into a smaller crop production area. Experience has shown, however, that crops do not necessarily respond to exaggerated applications of green manures. There are few farming systems in which green manuring is economically feasible (43,46) since land is tied up without immediate economic return. Where green manuring is practiced, 5 tons of green matter per hectare is an accepted application rate (47). This would represent an addition of only 40 kg/ha of nitrogen to the soil, of which only about 20 kg would mineralize to the benefit of the crop. Read data, though limited, supports such an estimate. Green gram contributed 22 kg of nitrogen to following crops and calapo/stylo green manure contributed 15 kg (48).

Moving on to consider the nitrogen economy of mixed cropping systems in which the legume and non-legume are growing concurrently, the situation is more complex. Legumes usually take up less soil nitrogen in competition with non-legumesand a greater fraction of the nitrogen they accumulate in mixed crop is fromfixed nitrogen. Somewhat suprisingly, the nitrogen fixation of intercropped beans (Phaseolus vulgaris) per hectare is not significantly different from beans raised in monoculture (49). This is attributed to competition between the maize and the bean for light and nutrients beginning after the decline in nitrogen fixing activity in the root nodules of the beans. Not all legumes shut down nodule function as early in the growth cycle as Phaseolus vulgaris and the effect of intercropping on nitrogen fixation may be detrimental in other intercropping systems.

It is a common misconception that there is a substantial direct transfer of nitrogen from the legume to a non-legume companion species in a mixed cropping system. There is no convincing evidence that actively growing, healthy legumes, whether grain or forage, excrete significant amounts of nitrogen from their roots or nodules.

The hypothesis originally proposed by Virtanen and co-workers (50,51,52) that surface excretion of simple amino compounds from healthy, functioning legume root-nodules resulted in direct transfer of significant quantities of nitrogen to non-legume companion species has found little support from other workers (53,54,55,56,57).

Subsequent research under carefully controlled conditions using the 'fog box' technique (58) indicated that excretion of a wide range of substances from plant roots does occur, but that the quantities involved are small, being less than 0.5% of the plant's nitrogen (59). Stated differently, a crop fixing 100 kg of nitrogen a year would excrete only 0.5 kg to the soil.

Nitrogen benefit to non-leguminous crops through association with companion legume species is considered to be of an indirect nature through loss and decay of shoot, root and nodule tissue, or by recycling via the grazing animal, rather than by a direct pathway (60,61,62,63).

Clearly then, mixed cropping systems that aim to utilize legume-fixed nitrogen for the benefit of a companion non-legume species must match species such that the non-legume is longer-lived than the legume because nitrogen will be released in significant amounts only after cessation of active growth and decomposition of tissues of the legume. The maize/bean association used widely in Latin America exemplified this principal. Estimates place fixation by beans at 20 - 40 kg of nitrogen per growing cycle (64). Assuming 70% removal of nitrogen as protein in the legume grain, this leaves only 6 - 12 kg in legume residues of which 3 - 6 kg (assuming 50% mineralization) will be mineralized in time to benefit the maize. Some estimates place the mineralization that can benefit a companion species as low as 20% and consistent with this it is not uncommon for there to be no detectable nitrogen benefit in companion crops that are intercropped with legumes.

From the preceeding quantitative considerations, it is clear that the biological nitrogen fixation benefit to non-legumes due to inclusion of legumes in a cropping system is small indeed compared to the level of nitrogenous fertilizer use in the more intensive cereal production systems of the developed world. Thus the principal contribution of biological nitrogen fixation to human nutrition will continue to be via the protein in legume grains. An important implication of the data discussed here is that any suggestion of substantial replacement of nitrogen fertilization of cereals and root crops by biologically fixed nitrogen is unrealistic as these crops are known to respond to levels of nitrogen fertilizer far in excess of those which could currently be supplied through BNF by legumes. Thus, there is an urgent need to devise ways of increasing the contribution which BNF by legumes can make to cropping systems as a complement to N fertilizer-based production rather than as an alternative to it.

Legumes can be managed to increase their nitrogen contribution to companion or following crops. They vary in total nitrogen fixed, the proportion retained in non-harvested residues, the percentage nitrogen level in residual tissue and the facility with which the organic nitrogen is mineralized. The greater these parameters, the greater will be the residual nitrogen benefit. Given this situation, the priority given in legume breeding programs to improving their harvest index, i.e. maximizing the fraction of each plant's total production that is removed as grain, should be called into question.

In summary it can be said that the principal benefits from BNF through the use of legumes in farming systems of the tropics are derived from the dietary protein of the legume grain, the multiple uses which legumes serve for the subsistence farmer, and the greater stability of yield and financial return of intercrops over monocrops. The indirect benefits from contribution of biologically fixed nitrogen to companion or following species are small but are significant at present in the context of input levels in subsistence farming in the tropics.

At present there is insufficient reliable data on the benefit to be derived from enhancing, through inoculant technology, the nitrogen fixation in tropical legumes over and above the level which would be anticipated from spontaneous nodulation with native strains already present in the soil to advocate that rhizobial inoculation always be performed. It is tempting to recommend rhizobial inoculation of all legume sowings as an insurance measure against the risk of nodulation failures that would otherwise occur. However, inoculant technology does represent a cost, albeit small, and does add a degree of complexity to the sowing practice. Thus inoculant technology should only be advocatd when there is known to be a need-to-inoculate and a demonstrable benefit therefrom. Additionally the concept and practice of inoculant technology is so foreign to farmers' normal practices that it should not be recommended lightly. A subsistence farmer can be forgiven for not comprehending nor accepting a technology that involves sticking black powder containing bacteria to his seeds. This contradicts concepts about which he has only recently become educated, namely, that bacteria are bad and clean seed is important. It is to be questioned whether inoculant technology in this form will ever be accepted widely among subsistence farmers in the tropics and subtropics. Further discussion of this topic follows later.

Unfortunately many trials performed to evaluate inoculant technology with tropical legumes under tropical conditions have been performed with imported inoculants which may not have contained acceptable levels of viable rhizobia at the time of use. Lack of reponse to inoculation in such trials does not preclude the possibility that the legume could potentially benefit from inoculation. More recently coordinated networks of trials have been initiated to determine whether there is an economic yield benefit from inoculation of legumes or not. INTSOY conducts International Soybean Rhizobium Inoculation Experiments (ISRIE) throughout the tropics. CIAT distributes an International Bean Inoculation Trial (IBIT) through Latin America. The University of Hawaii coordinates an International Network of Legume Inoculation Trials (INLIT) offered for 13 agriculturally important legumes and involving 3-stage experimental program in which cooperators throughout the tropics select strains specifically for their legume variety and local soil conditions thereby maximizing the opportunity for a yield response following inoculation.

Future potential of legume-based BNF technology

Despite their seeming attractiveness for sustained productivity for low-input production systems, and despite also their consistent strategic use in many farming systems of the tropics, legumes have remained minor crops in the systems in which they occur (65).

Why is this the case, and what factors would lead to greater use of nitrogen fixed biologically by legumes? A small-scale, subsistence farmer elects to raise those crops that best meet the various needs of his household but also chooses one crop, at least, to sell or exchange for goods or services for which he is dependent on others. Large-scale farmers consider primarily the economic return and facility of management associated with the crops they will choose to plant. A grower preference

for cereals over legumes, when the grain is to be marketed, would be understandable. It is usual for yields of cereal grains to be as much as four times higher than legumes (typically 3.0 t/ha vs. 0.7 t/ha). Although the protein content of legumes is much higher (30%) than cereals (6%) the market value of legume grains, albeit higher than for cereals, does not compensate the grower for their low relative yield.

Many factors will contribute to an increase in the use of legumes. Cereals will continue to be the major source of protein and calories for human nutrition on a worldwide basis but an increase in importance of root and tuber crops and plantains over the next two or three decades is anticipated (66). Legumes can be expected to be one means of complementing the dietary quality of these starchy foods that are deficient in protein.

Another factor that has already caused a re-appraisal of biological nitrogen fixation through legumes is the cost and availability of energy for production of nitrogen fertilizers. Already 20% of N fertilizer production in the U.S. is cost-ineffective because of the cost of energy (in the form of natural gas) for the process. Producer costs have been calculated as $160 per ton (67) whereas the selling price is in the range of $85 - $105 per ton. It is predicted that by as early as 1984, 60% of the world's ammonia production will be by the USSR, eastern Europe, and the developing countries, and that ultimately N-fertilizer production will be economically feasible only in those countries with huge surpluses of natural gas (mainly Mexico and Indonesia).

Thus biological nitrogen fixation through the use of legumes may be resorted to increasingly, not only to reduce the cost of on-farm inputs, but also to save foreign exchange and avoid over-dependence on foreign powers.

But economic pressure alone will not guarantee adoption of biological nitrogen fixation-based technology without compelling demonstration of greater benefits from BNF since the energy crises of 1973/4 and 1979 has brought it under the scrutiny of agencies and individuals whose concern is its viability as a productive agricultural technology now rather than its often acclaimed potentials for the future. This should encourage the agricultural research community to undertake a comprehensive program of technology development in which the relative distribution of funding and manpower investment is realistically prioritized. Thus, research aimed at stabilizing grain legume yields is likely to increase the contribution of biological nitrogen fixation in tropical farming systems to a greater extent than much of the research on the BNF process per se in grain legumes. Similarly, research involving selection of forage legume germplasm that is adapted to the soils and climates of the world's under-utilized savannahs, and development of appropriate legume-based pasture management and utilization technology can be expected to bring about a major increase in the use of biological nitrogen fixation even without further research on the BNF process per se in those legumes. These statements assume, of course, that those BNF components of each legume production package that are acknowledged to be indispensable can be guaranteed, i.e. effective nodulation. Since this is not always the case, those specific aspects of BNF research which tackle the factors which limit

nodulation and nitrogen fixation by legumes in tropical soils should be singled out and given highest priority. Unless pursued concurrently and with an intensity that assures that outputs on the BNF component for each legume are in synchrony with other advances in the use of those legumes, performance of the legume "package" will be seriously impaired.

Constraints to implementation of BNF technology

There are still many unknowns in the scientific understanding of BNF, and research into the biochemistry and genetics of the process is particularly intense and competitive.

But few, if any, of these unknowns are really constraining the implementation of legume-based BNF technology. The basic principles of inoculant technology have been known for many years and have already made major contributions to agricultural production initially in Australia and, more recently, worldwide as soybean cultivation has been increasing. The real constraints to fuller implementation of BNF technology relate to delivery of the technology, both to potential inoculant producers and to farmers, and acceptability of the technology, again by both the inoculant producers and farmers.

There has not been adequate demonstration, under realistic, farm-relevant conditions in the developing countries, of the yield increases and/or reduced fertilizer needs that are repeatedly stated to be the benefits of BNF technology. In some cases, inoculation trials have been performed and no response obtained. But these trials have been mainly with imported inoculants, the quality of which at the time of their use was not or could not be verified. Thus a related constraint is the lack of trained personnel with the essential combination of agronomic and microbiological skills for executing production-oriented research on BNF technology including development of appropriate Rhizobium strains and inoculation procedures for use under the variety of circumstances encountered in the tropics and subtropics. It must be remembered that current inoculation technology as used in the U.S. and Australia is suited to legumes grown under favorable conditions with relatively high complementary agronomic inputs. Transferability of this technology to situations where the legumes are grown usually in marginal conditions with minimal inputs, and confronted with one or more soil and climatic stresses is in some doubt (68).

It is the genotype of the legume which is to be inoculated that is the prime determinant of the strain used in rhizobial inoculants, rather than the characteristics of the soil into which the inoculant will be introduced. This is contrary to what is expected by many first-time users of inoculants.

For example, in providing inoculant services in Latin America and Hawaii, it has been common to receive data from soil analysis together with requests for inoculants. This illustrates that farmers expect that the selection of legume inoculant is made after consideration of local soil and climate, just as would be the choice of crop variety. Yet there is only

one instance in which an inoculant strain recommendation for an inoculant in commercial production takes into consideration the soil characteristics into which it will be introduced. Rhizobium strain CB 81 is recommended for Leucaena leucocephala sown in acid soil and NGR 8 for alkaline soils (69).

When soil characteristics are very different, the response to inoculation and the relative performance of rhizobial strains is also different. Even when soils were selected for their apparent similarity, the performance of rhizobia in those soils was very different. It is not suprising therefore that some authors have advocated that simple "need-to-inoculate" trials always be performed at the local level due to the unpredictability of the response to inoculation (16,18,32). If carried to its conclusion, this suggestion would result in legume inoculation being tested, essentially by trial-and-error, at every site where legumes are to be grown. It is certainly to be hoped that inoculation technology is more transferable than this, otherwise its value as an agrotechnology is questionable.

It is well known that there are significant differences between sites in the size of their indigenous rhizobial populations (70,71) and in the range of strains of Rhizobium in the indigenous microflora (71,72). Such differences have been attributed to the effect of soil factors (72,73) though the possibility of widespread correlations between specific soil characteristics and rhizobial occurrence in tropical soils has not been critically examined.

It is also acknowledged that the response by tropical legumes to inoculation with rhizobia varies from site to site (15,16,32,,74,75). Such variation has been attributed to: differences in number, effectiveness and competitiveness of native strains (17,71,76,77); variation in quality of the inoculant at its time of use (78); and to variation in soil nitrate levels (79). The possibility that the response to inoculation could be predicted on the basis of a more thorough description of soil and environmental characteristics has not been tested.

It is also known that the relative performance of strains selected under optimal conditions for a specific legume is variable, depending on the site to which they are introduced (15). With inoculants that contain a mixture of strains of Rhizobium it is common for one strain to dominate in the nodule population that results from the inoculation (80,81). The possibility that rhizobial strains might be selected for adaptation to particular soil and environmental conditions is not presently exploited in tropical agriculture.

A serious constraint to fuller implementation of BNF technology is non-availability of domestically-produced, high quality inoculants within each of the countries of the tropics and subtropics. Thus factors which deter government organizations or private enterprise from undertaking inoculant production in a particular country are also constraining BNF technology. Among these are: high capital cost of inoculant production plant (of type used in the U.S. and mistakenly assumed to be a prerequisite for any production plant); high operational cost associated with retaining a professional and well-trained staff to run the plant; operational risks

associated with losses due to such factors as contamination; absence in most developing countries of an adequate infrastructure that would permit marketing and distribution of a biological production with notorious vulnerability to damage by high temperatures; reticence to embark on an enterprise in advance of official control standards being established (confounded by official reticence to set standards until there is an industry to be controlled); insufficient present demand and uncertain future demand for inoculants.

As has been referred to already, the present nature of the technology meets considerable farmer resistence, i.e. the coating of seeds with peat inoculant. In Brazil, packets of inoculant are included "free" by some seed distributors with all seed sales. However, the inoculant is frequently discarded by farmers not only because of the nuisance associated with use of the inoculation on a field scale, but also in part because of an unfortunate impression that if inoculant is "free" it is suspect and of little value.

The cost of inoculants is not usually a constraint to the use of inoculant by farmers who outlay capital for seed. Inoculant will seldom exceed 1% of the seed cost. For subsistence farmers who do not ordinarily purchase seed off-the-farm, the capital outlay for inoculant, albeit small, may be a disincentive to the use of inoculants. Cost becomes a more important consideration with granular forms of inoculant in which the rate of application is much greater than with seed-applied inoculant.

BNF technology is a difficult technology to deliver by normal extension mechanisms. Thus a lack of illustrative and explanatory pamphlets and other aids both for extension agents and the farmers with whom they have contact is also a constraint to implementation of BNF technology at the farm level.

Furthermore, few of the senior administrators and decision makers who determine agricultural policy in developing countries are fully aware of the applications for legume-based BNF technology in the agriculture sector of their countries. Most policy makers are aware of some of the attributes of legumes. Relatively few of those individuals appreciate the role played by biological nitrogen fixation in legumes and among those few an even smaller proportion recognizes that it may be essential to employ specific technologies to ensure that nitrogen fixation occurs at all, let alone at a maximal rate. Thus there is a need for educational material, specifically developed for this clientele group, bringing to their attention the real need to adapt currently available technology to the particular circumstances in which it is to be employed in their country.

As BNF technology is being implemented, new constraints are emerging that are best described as "scientific" and are researchable. For example, some countries do not have peat deposits suitable for carrier materials for inoculant production and alternate materials must be identified and validated. Also, specific soil and climatological stresses such as extreme soil acidity and the associated high levels of toxic elements like aluminum and manganese may require selection of strains of rhizobia tolerant to those conditions for use in inoculants.

It could be suggested that the large number of researchers with the competencies required for BNF research but who expend their energies and resources researching aspects of BNF other than limiting factors such as the examples cited above is also a constraint to fuller implementation of BNF technology. Funding agencies do not always recognize a distinction between applied and less useful research that competes for resources under the general subject matter area of biological nitrogen fixation. Biological nitrogen fixation has great pertinence to agricultural production in developing countries but not all research conducted under the BNF umbrella is applicable in agriculture.

Scenario for full implementation of BNF technology

Given that the constraints to fuller implementation of BNF technology are not solely scientific but include cultural, socio-economic and political factors, the scenario in which BNF might realize its potential would necessarily be multi-faceted and comprehensive.

The current trend toward energy-efficient farming systems to reduce capital outlay for fertilizers that must be imported can be expected to continue and intensify. Because of the high energy consumption during manufacture of nitrogen fertilizers, their price and availability is influenced increasingly by oil-rich nations. There is added attractiveness in alternate nitrogen sources to avoid even further dependence on foreign powers. Legume-based BNF technology is the major option available and is likely to be resorted to more and more. As has been described, the use of legumes and appropriate inoculant technology has the potential to increase the amount of biologically-fixed nitrogen entering agricultural production systems. Given that the main value of legumes is their high-protein grain, rather than their nitrogen contribution to non-leguminous food crops such as cereals and root crops, the scenario for full realization of BNF technology's potential would need to include a swing in consumer preferences away from crops that depend so heavily on nitrogen fertilizer. Thus, in the gambit of BNF research priorities, attention will need to be given to learning the cultural and scientific bases for these preferences and to alleviating, where possible, the constraints to greater consumer acceptance of legumes.

As explained earlier in this report, the major increases in benefits from legume-based BNF technology will arise through: an increase in the total acreage in legume production; innovative use of legumes in roles they have not previously occupied; and by ensuring that biological nitrogen fixation in those legumes is maximal through appropriate inoculation technology. Much remains to be done to improve the biological nitrogen fixation components in the technology package for legumes in the role they currently play in agriculture. There is a wide discrepency between farmers' yields and the known yield potential of the grain legumes. Furthermore, it is disconcerting that in the majority of reports of legume trials that included rates of nitrogen fertilizer application, the legumes responded to nitrogen ferilization -- disconcerting because this means that even when legumes were grown under favorable management in experiment stations, let alone in farmers' fields, the symbiotic association of the legume with

Rhizobium was defective. There is therefore the potential to double or triple the nitrogen benefits described in this report through development of technology that would assure establishment of maximally effective rhizobial symbioses in tropical legumes under tropical conditions.

Greatest future potential would appear to rest in the development of: legume-based pastures and viable multiple-cropping systems including legumes for under-utilized savannahs; agroforestry systems that combine fast-growing, nitrogen-fixing trees, leguminous and other crops to meet the food and fuel requirements of the rural poor; fast-growing leguminous trees for reclothing water catchment areas following forest clearing; legume-based cropping systems to give sustained productivity in tropical soils following jungle clearing which typically exhibit a rapid decline in fertility under conventional cropping; selection of deep-rooted, drought-tolerant leguminous trees that can serve as browse species in the world's dry lands.

Reference has already been made to the need to exploit fully the variation in host plant x rhizobial strain x environment interaction in selection of the optimal BNF package for each circumstance. This would require that all current legume programs retain the services of a trained professional microbiologist, a suggestion that is just not practical. Firstly, few legume programs can afford the luxury of a full-time microbiology position and secondly, there is currently a worldwide shortage of professional soil microbiologists that is unlikely to be alleviated significantly for about ten years. The world's major multidisciplinary legume programs should, however, have their own microbiologist. This is already the case with the IARC programs for beans, cowpeas, pigeon peas, groundnuts, chickpea, and the tropical forages. INTSOY, working with soybean, has its own soil microbiologist. Also there are several national legume programs into which microbiological support is intergrated through a participating institute with expertise in the BNF area (e.g., Brazil, India).

The needs of the multitude of other legume programs for BNF expertise could be met through the provision of one (or more) BNF Resource Center(s) established to provide technical assistance, offer support services (germplasm and information), provide professional and technical training, and conduct such research as may be necessary for adaptation of BNF technology to specific local conditions but which is beyond the capability of local researchers. Such centers would require a critical mass of BNF researchers to be able to carry out a comprehensive support program and still retain a capability to respond to technical assistance requests that are sometimes difficult to anticipate. The BNF Resource Center(s) would best be located at universities in developed countries and preferably in the tropics. Location of a BNF Resource Center at a university facilitates provision of professional training, an important output for assuring that ultimately national institutions in developing countries can sustain their own BNF programs. Short-term, non-degree training programs in BNF technology should be offered to key personnel responsible for providing bacteriological backstopping to production-oriented research programs involving the legume/Rhizobium symbiosis. This is more effective in the short-term than Ph.D. or M.S. programs which tend to be a passport out of research into better paid administrative positions for many graduates

returning to their home country. The short courses should be mounted in cooperation with developing country institutes to generate a regional capability for offering such courses. They should be complemented by on-the-job training tailored to the needs of selected individuals that would be conducted at the BNF Resource Center and include visits to pertinent industry facilities.

Such BNF Resource Centers would engage information specialists for development of communications materials suitable for the many clientele groups to whom BNF technology has to be delivered. This would range from newsletters for administrators to pamphlets for extension agents and include updating developing country researchers, who often do not have access to libraries, on progress in BNF research.

There has been an increased tendency in agricultural research to focus manpower and resources on improvement of single commodities. Thus the IARCs are characterized by multidisciplinary teams with specific crop and/or geographic mandates. Similarly in the CRSP activities under Title XII a commodity approach has emerged. The suggestion for establishment of a BNF Resource Center would be considered by some as going against this trend and viewed as a return to discipline-oriented research. This author contends that the key element in the acknowledged success of commodity programs such as some of those in the IARCs has been that they are highly focused and actively managed in pursuit of well-defined research priorities rather than attributable to the commodity approach per se. A program investing manpower and financial resources in an actively managed, carefully prioritized BNF program sharply focused on the constraints to full implementation of BNF technology can be expected to make real progress toward that end. The specialized and sophisticated nature of *Rhizobium* bacteriological expertise and the scarcity of experienced manpower is further justification for assembling a critical mass of rhizobiologists in a single BNF Resource Center.

An additional advantage to the existence of such a BNF Resource Center would be a capability to extend BNF technology developed at a particular IARC to crops and regions outside the crop and geographic mandate of that IARC but in which there is a strong likelihood that the technology is immediately utilizable. Staff of the BNF Resource Centers would travel as required and undertake short (1-2 months) or longer (3 months - 3 years) assignments in support of specific outreach activities when warranted. Conceptually the personnel of the BNF Resource Center would have their attention drawn to researchable constraints on BNF technology in real agricultural situations in the developing countries when interacting with local cooperators during duty travel. The BNF Resource Center would work closely with other universities and research organizations to which specific research on factors limiting BNF utilization could be referred under sub-contract when those institutions are recognized centers of excellence in the pertinent sub-discipline and have a comparative advantage to provide the required support.

The BNF Resource Center would need to develop links with commercial inoculant producers to mount appropriate assistance programs for government organizations or private enterprise in developing countries contemplating

inoculant production. Such programs would cover not only technical aspects of the production of inoculants but also the business aspects of small enterprise production, marketing and distribution of inoculants. The BNF Resource Center should develop specifications, including sources of all equipment items, for inoculant production facilities that would be feasible at levels of capital investments ranging from $50,000 to as high as $1 million. The Center should also advise governments on an appropriate mechanism for quality control.

The Center would also need to develop strong links with major legume germplasm centers and those involved in legume improvement to encourage simultaneous exploitation of host legume and rhizobial germplasm in selections for particular soils and climates.

The BNF Resource Center would take a major organizational responsibility for calling such workshops and scientific meetings as may be necessary to coordinate international experimentation and provide for dissemination of results.

The major activity to be undertaken by the BNF Resource Center would be the coordination of a competently executed sequence of standardized experiments designed to generate the data necessary to quantify the economic yield benefit attributable to legume inoculation under field conditions. Such trials would also serve as local demonstrations of the benefits from legume inoculation.

The core budget for such a BNF Resource Center should be guaranteed by the host government through its agency responsible for international development. The host institution (university) cannot realistically be expected to provide direct financial support for such a Center given that the Center staff will not have substantial conventional instructional responsibility and that the research output will aid mainly foreign nations with only minor spin-off benefits for agriculture in the state or province where the Center is located. The mandate of a BNF Resource Center is international and therefore the support should be international.

There is understandable reticence on the part of international funding agencies to expend resources in a center located in a developed country.

This author contends that it is in the best interests of the developing countries that they be supported in their BNF programs by a Center located in the tropics but sited in a developed country where it can receive unimpeded logistic support for its sophisticated operations and enjoy continuity of service from high caliber professional staff. Such a Center would be ultimately more cost-effective than fragmented support to a myriad of in-country programs, an approach that often causes wasteful duplication of effort. Furthermore, support of a BNF Resource Center, for example, in the U.S. with funding by USAID, would be prudent use of public funds. A maximum share of the Center budget would be expended in the U.S. sustaining employment of the U.S. residents and strengthening a U.S. institution without lessening the level of development support for the developing countries. Additionally, a greater degree of control could be exercised over the activities of a U.S.-based Center than is possible with grants to foreign institutions.

Agencies that could be anticipated to be in the portfolio of contributors to a BNF Resource Center would be: FAO, UNEP, Unesco, and UNDP. Technical assistance on a continuing basis to any specific country ought to be funded externally as a special project with funding arranged by that country drawing on whatever national budget and international development assistance grants or loans are accessible to it.

Impacts of wide-scale implementation of BNF technology

The use of legumes to benefit other non-leguminous crops implies mixtures or rotations of crops. The production systems are therefore labor intensive and, in the case of mixed crops, not readily mechanized. Since this is already the circumstance of most production in developing countires, no impact is likely. If BNF technology were to be employed in the U.S., for example, this would necessitate a swing away from the highly-mechanized monocrop systems currently employed and a major demand for farm labor.

Inoculant technology is scale-neutral but the use of legumes as green manures or in multiple crops and rotations is not. Such practices are more suited to small farms than large farms.

Inoculant technology does not increase nor displace appreciably the demand for labor. The intentional use of legume residues for the benefit of non-leguminous crops will create demand for laborers, since the technology involves extra cultivation steps that are not easily mechanized.

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APPENDIX E - TECHNICAL REVIEW

Revised:4/29/86: Revised 5/8/86
DIMPEX

Technical Strategy

The question is no longer "if we can engineer beneficial organisms", but "what we should engineer them for". The modern revolution in molecular biology has made it possible to alter the genetic composition of microorganisms and bestow upon them attributes desirable to man. But, our lack of knowledge of how the new organisms would perform in the field, has made it difficult to tailor organism to order. The challenge is to be able to match the genetic potential of the organism to the controlling parameters of the environment.

In the proposed project, we have taken a multi-tier, techno-social approach to wider utilization of agricultural biotechnologies in developing countries of the tropics. In Program 1, (Genetic Technologies) we propose to develop superior symbiotic systems through modern and conventional genetic techniques. However, even assuming the availability of "super-strains", whether through selection or genetic engineering, there still remains a major technical stumbling block to wider utilization of the superior product: the physiochemical and biological barriers of the soil that would prevent the establishment and optimal performance of the engineered organism. To overcome these barriers we propose Program 3 (Environmental Constraints) that will allow us to alleviate the environmental stresses that most limit establishment of desired strains.

Implementation of useful agro-technologies in the developing world is, perhaps, even more limited by product availability and an effective distribution system. We submit that lack of incentive is the real cause of this constraint. To motivate potential LDC entrepreneurs, we will offer assistance in technical and business procedures with the aim of promoting formation of small businesses and manufacturing plants.

Program 1. Genetic Technologies

Major advances in molecular biology have provided new techniques that allow us to control and manipulate the genetic material of organisms to an extent previously deemed impossible.

It is a routine procedure now to mobilize plasmids carrying specifc information from one organism to another (for details, Cold Spring Harbor Manual "Molecular Cloning"). For example, in *Rhizobium* intra-and-interspecific transfer of symbiotic plasmids has been accomplished by several groups (for example, Beringer *et al.*, 1978: Pilacinski and Schmidt, 1981; Stanley and Dunican, 1979). Genomic and plasmid libraries of several inoculum strains of *Rhizobium* have been constructed and many specific genes identified (for review see Schwinghamer, 1977).

Studies of the genetics of nitrogen fixation and host infection have almost exclusively been done with the fast-growing species of temperate legumes such as peas, beans, clover and alfalfa (for further discussion see

'Sadowsky and Bohlool, 1983). In these rhizobia, the genes that code for the nitrogenase enzyme and some of the genes for host-recognition and infection are located on plasmids (for example, seeds that can be easily mobilized). Very little, however, has been done with the genetics of other groups of rhizobia, especially with those that nodulate tropical food and fuelwood legumes. Program 1 is directed at improving the effectiveness of symbiotic nitrogen fixation through genetic manipulation of the bacterial partner, Rhizobium, and through biotechnological approaches to the modification of the host legume. We propose to : 1) use the genetic library already present in the resident rhizobia of tropical soils, adapted to the prevailing stresses, and engineer into them genes for better nodulation and/or higher levels of nitrogen fixation. It is possible now, through molecular cloning and recombination, to construct plasmids that contain multiple copies of nitrogen fixation (nif-) and host infection (inf-) genes. Through collaborative efforts with LDC scientists involved in related research (Dr. R. Palacios, Mexico, Dr. K. Malik, Pakistan, and Dr. N. Boonkerd, Thailand) we will develop and test rhizobial strains that harbor multiple copies of Nif-structural genes and inf-genes for nodulation of specific tropical legumes; 2) identify genes involved in inter-strain competition in organisms already recognized as super-competitors (e.g., TAL 1145, CIAT 899, Hawaii 5-0, USDA 123) and introduce these genes into efficient inoculum strains for other tropical legumes. Through transpon (Tn-5) labeling of competition genes, we will generate mutants. Theses mutants will be sent to our collaborators in Mexico, who will isolate and clone the genes, which we will then introduce into different species of tropical rhizobia and test for performance in competitiveness under stressful conditions; 3) utilizing the diversity of tropical environments available to NifTAL, in Hawaii and through NifTAL's network (INLIT) cooperators we will test performance of tissue culture generated crop and tree legumes; 4) through EMS-mutagenesis breeding we will develop mutants of food and tree legumes for production and research. There has recently been exciting developments with soybeans and peas through mutagenesis selection. Supernodulators that continue to function optimally even in the presence of high levels of soil N have been developed (10). Similar procedures will be used to select improved lines of several tropical legumes. Non-nodulating (non-nod) characteristics can also be generated through mutation breeding. Non-nod genes have already been identified in soybeans and moved around into other cultivars, through conventional breeding, to develop non-nod isolines of important soybean lines. These are extremely valuable in quantification of BNF. Quantification, of course is very difficult with fuelwood and forest legumes. Through mutgenesis selection we will develop non-nod isolines of Leucaena for experimental purposes.

Winged bean (Psophocarpus tetragonolobus) has been hailed as "the soybean of the tropics". But, its climbing habit deters from its usefulness for wide-spread utilization. We will select, through mutagenesis breeding, erectoid lines of winged beans.

The methodology for mutagenesis selection would be essentially that used for peas and soybean (10). Imbibed seeds will be soaked for several hours in a solution of EMS (Ethyl Methane Sulfonate) to affect mutation. Seeds will be planted in soils depleted of nitrogen by Bagasse incorporation.

•Flowers will be labeled and allowed to self-pollinate. Seeds from these lines (M1) will be planted out. Mutations will be expressed at this level. Mutant lines with expressed phenotypes (supernods, errectoid, non-nods) will be allowed to self pollinate. Seeds from these lines (m2) will be used for further testing at NifTAL and, through the network of collaborators, at other tropical locations.

Program 2. Development of Methodologies

Effective biotechnologies rely heavily on advanced techniques in biology, biochemistry and engineering. We will develop new, and/or refine already existing, methods used in BNF research.

Seriological techniques are tools for identification of specific microorganisms used in research, production and agricultural practices. Polyclonal antibodies provide means for fingerprinting of the total antigenic composition of microbes. They have been useo extensively in characterization and monitoring of Rhizobium. NifTAL will continue to provide this service for research and education, as well as for quality control of industrial Rhizobium inoculum.

In some groups of Rhizobium, antigenic similarity among strains preclude the use of polyclonal antibodies. We will develop monoclonal antibodies for select groups of tropical rhizobia for research and training purposes. In many LDC institutions where serological tools, equipment and expertise are not readily available, inoculum strains can be labeled with specific antibiotic markers to facilitate their easy detection and identification. At NifTAL we will generate such marked strains to be used by LDC research and production collaborators.

Program 3. Environmental Constraints

During the nine years of its existence, NifTAL has established a functional network of collaborators and field sites throughout the tropics, to test the benefit of legume inoculation with Rhizobium in LDC's. Former NifTAL trainees and other affiliated scientists have carried out INLIT (International Network of Legume Inoculation trials) at 200 overseas sites in 53 countries in Asia, Africa, Latin America and the U.S.

The INLIT experience has been extremely valuable. First and foremost is the establishment and understanding of the infrastructure of a functional network in LDC's. Equally significant are: 1) identification of reliable and capable scientists throughout the developing world; 2) identification of institutional capabilities in several LDC's; and 3) demonstration of benefits of inoculating legumes at numerous sites.

While the INLIT has provided useful information on which legumes respond to inoculation at different field sites in the tropics, the experiments were not designed in such a way as to define the major ecological factors which control legume response to inoculation with rhizobia. Explaining the response to inoculation in ecological terms is necessary in order that a

predictive model can be developed. This model can be utilized by planning institutions and extension workers in their efforts to coordinate the production, distribution, and application of complimentary agricultural inputs.

Factors affecting the response to inoculation by a specific legume may be due to soil physio-chemical constraints (i.e., soil nitrogen, pH, fertility, etc.) or biological constraints (indigenous populations of rhizobia and/or other microorganism and soil invertebrates). These specific Rhizobium responses to soil ecological factors will be similar or have a great deal in common with the response of other microorganisms introduced into a soil environment. Therefore, data obtained from experiments in this proposal will have relevance for microbial ecology in general and of possible usefulness to scientists contemplating the introduction of genetically engineered organisms into soil environments.

Program 4. Regional Resource Centers

Regionalization of Agrotechnology Resources: to tailor development support services offered globally from NifTAL, Hawaii, to specific regional needs. A regional Resource Center provides a stong foundation for research, training, technology transfer, technical assistance, as well as information dissemination. Because such a center is oriented to the needs of a specific region, strong links can be established with other institutions which are engaged in Agricultural development work.

Many of the technical resources for development assembled during NifTAL's early years remain widely applicable throughout the tropics. But, as the emphasis at NifTAL has progressed from legume inoculation into the stategic use of legumes in tropical cropping systems, it has become necessary to tailor NifTAL outputs to the vastly differing practices of the world's major geographic regions and the contrasting degree of advancement of agriculture in specific countries.

NifTAL hopes to contribute ultimately to establishment of a comprehensive BNF capability in the national agencies of each developing country. Establishment of regional BNF Resource Centers is viewed as an interim step toward that goal.

This step also makes it easier for USAID's Regional Bureaus and Country Missions to tap NifTAL resources in support of regional and country programs involving legume crops.

Bangkok was considered the most appropriate location for the Regional Resource Center because of Thailand's pivotal position in the region, and the longstanding tradition of cooperation between the University of Hawaii NifTAL Project and the Soil Science Division of the Thailand Department of Agriculture. The Department of Agriculture endorsed the BNF Resource concept and, under a May 1983 Agreement, offered to host the Center at the Rhizobium Building, Bangkhen, Bangkok.

In October 1983, a planning workshop was held in Bangkok to derive a workplan for the BNF Resource Center. Country representatives detailed the most pressing research and promotional needs to put BNF to work in their farmer's fields in the client countries of the region (Pakistan, India, Sri Lanka, Nepal, Bangladesh, Burma, Thailand, Indonesia and the Philippines).

Anticipated goals of the BNF Resource Center Program are as follows: 1. Strengthen Bangkok Center and establish addional ones in the Near East, Africa, and Latin America; 2. Publish quarterly Newsletters, initially for each region and within 2-3 years a consolidated edition for all centers; 3. Assist regional/national seed projects for food, oil, and agroforestry purposes; 4. Design and implement production and distribution systems; 5. Assist in establishment and operation of small businesses for production and/or marketing of inoculum; 6. Provide to area scientists selected rhizoial cultures and inoculants which are adapted to regional soil constraints; 7. Promote collaborations among regional scientists by engaging them in the INLIT Program (International Network for Legume Inoculation Trials); 8. Link regional scientists through workshops, information, document exchange, and the newsletter; 9. Assist AID Missions with Project design and implementation.

In order to implement the program, the following input are necessary: 1. One senior scientist (Ph.D.) outposted in each center as Center Director; 2. One M.S. or B.S. scientist from the region as Assistant Director; 3. Research Assistants; 4. Clerical and information specialist; 5. Marketing specialist and accountant; 6. Rental of office, laboratory and field facilities (if not contributed by host institution); 7. Equipment for office, laboratory and field operations; 8. Travel budget; 9. Operational funds; 10. Back up support at NifTAL Headquarters.

The BNFRC in Bangkok receives considerable support in the form of facilities and personnel from the Thailand Department of Agriculture. NifTAL estimates that approximately $250,000 from its core budget is expended each year on direct support in Thailand and on back-up costs at headquarters. A new center in North Africa or India is estimated to require about $500,000 per year for inputs. This estimate includes the costs of facilities, vehicles and equipment, amortized over a 5-year period.

Program 5. Small Business Promotion

Successful utilization of modern agrotechnologies by farmers has been achieved in the U.S. and other developed countries (DC's) mostly by creating an incentive system that benefits not only the user, but also the producer and distributor. Over the past 50 years in this country, USDA and the Land Grant Universities, have developed a solid base of knowledge and trained personel for BNF Biotechnologies. However, it was not until U.S. private enterprise recognized the commercial reality of this biotechnology, that Rhizobium became a widespread practice at the farm level. The experience of NifTAL and other BNF programs in LDC's has led to the conclusion that incorporation of BNF technologies into small farmer practices will take a multi-sector developmental and promotional approach.

The developmental phase involves: 1) Research to modify the technology to make it more adaptable for tropical conditions and appropriate for LDC applications and 2) Training to produce a cadre of personnel capable of applying the technology. The promotional approach must include information dissemination and successful demonstration of the benefit of the technology. This can best be accomplished through entrepreneurs who become motivated to promote the product and assure its success. To date at NifTAL, we have dealt primarily with training and education of national scientists, extension agents, and policy makers. Recognizing the power of private enterprise to promote and market new technologies, we propose a program to assist LDC entrepreneurs to establish small biotechnology businesses to produce, promote and distribute the product.

A recent experience at NifTAL serves to illustrate one possible model that would work in some LDC's. A former NifTAL trainee from the University of Ankara, Turkey, Dr. Vellitin Gurgun, has been successful in convincing some Turkish financiers of the commercial potential of Rhizobium inoculum. They have been able to raise enough capital through private investments and loans to be able to construct a production plant and have now offered to contract NifTAL, on a private basis, to assist in assembling fermentor units and providing the training necessary for production, packaging and dissemination. While this model might be practical in some countries we recognize the need for extreme flexibility when dealing in such a diverse array of political and economic systems. We submit that one or more of the models listed below could be made operational in the majority of LDC's that NifTAL has dealt with over the past 10 years.

Models for Small Companies in LDC's

Model	Description	Ownership & Operation % of effort LDC	DC	Example
1	Wholly-owned by LDC	100	0	(Turkey) Philippines, Thailand, Egypt, etc.
2	Mostly-owned by LDC	75	25	Colombia, Indonesia, Mexico, Costa Rica, Egypt
3	Mostly owned by DC	25	75	Ecuador, Boliva, Peru, Guatemala, Sri Lanka, Bangladesh, Pakistan
4	Franchise system owned by LDC or DC	100 (0)	0 (100)	China, Malaysia, India, Taiwan, Chile, Venezuela, Saudi Arabia, Uruguay

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APPENDIX F - MISSIONS, REGIONAL BUREAUS AND AID/W FUNDING

Revised: 4/29/86: Revised 5/8/86
DIMPEX

Improved Biological Nitrogen Fixation through Biotechnologies
Mission, Regional Bureaus and AID/W Funding

(In thousands)

Description	First Five Years						Second Five Years						Ten Year Total
	FY 1987	FY 1988	FY 1989	FY 1990	FY 1991	Five Year Total	FY 1992	FY 1993	FY 1994	FY 1995	FY 1996	Five Year Total	
Regional Resource Centers													
Asia and Near East													
Thailand	$ 250	$ 250	$ 250	$ 150	$ 100	$1,000	$ 100	$ 100	$ 50	$ 50	$ -0-	$ 300	$ 1,300
Morocco	250	250	250	250	250	1,250	150	125	75	50	50	450	1,700
Total ANE	$ 500	$ 500	$ 500	$ 400	$ 350	$2,250	$ 250	$ 225	$ 125	$ 100	$ 50	$ 750	$ 3,000
Africa - Senegal	$ 250	$ 250	$ 250	$ 250	$ 250	$1,250	$ 150	$ 100	$ 50	$ 50	$ 50	$ 400	$ 1,650
Latin America and Caribbean													
ROCAP	$ -0-	$ -0-	$ 250	$ 250	$ 250	$ 750	$ 250	$ 200	$ 100	$ 100	$ 50	$ 700	$ 1,450
Total Centers	$ 750	$ 750	$1,000	$ 900	$ 850	$4,250	$ 650	$ 525	$ 275	$ 250	$ 150	$1,850	$ 6,100
Short-Term Technical Assistance													
Asia and Near East	$ 200	$ 200	$ 200	$ 200	$ 200	$1,000	$ 200	$ 200	$ 150	$ 100	$ 100	$ 750	$ 1,750
Africa	50	100	100	150	150	$ 550	150	150	150	150	125	725	1,275
Latin America and Caribbean	50	75	100	100	150	475	150	150	150	125	125	700	1,175
Total Short-Term TA	$ 300	$ 375	$ 400	$ 450	$ 500	$2,025	$ 500	$ 500	$ 450	$ 375	$ 375	$2,175	$ 4,200
Grand Total	$1,050	$1,125	$1,400	$1,350	$1,350	$6,275	$1,150	$1,025	$ 725	$ 625	$ 500	$4,025	$10,300

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Appendix F
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APPENDIX G - NETWORK NARRATIVE AND COOPERATORS

Revised:4/29/86: Revised 5/8/86
DIMPEX

Improved Biological Nitrogen Fixation through Biotechnology
International Network of Cooperators

Networking is an important aspect of the Nitrogen Fixation by Tropical Agricultural Legumes (NifTAL) programs. By virtue of NifTAL's location within the College of Tropical Agriculture and Human Resources, Department of Agronomy and Soil Science, University of Hawaii, it has established working relationships with other academic departments located on the main campus and can draw on the vast resources available at the University. The University staff of scientists and experts who are available to NifTAL at no cost to the project are listed in Appendix J.

A biological nitrogen fixation network has been established to promote the use of nitrogen fixing microorganisms to increase crop production and reduce the dependency on nitrogen fertilizer. The purpose is to maximize the cost-effective transfer of technology from NifTAL and other donors where techniques are developed to LDCs in need of appropriate technical assistance. The network is also valuable as a way for exchanging successful practices among the scientists.

To achieve this purpose, NifTAL has used an integrated approach involving research, outreach and training activities. Through research, a comprehensive collection of rhizobium germplasm containing effective strains adapted to tropical conditions are available and are distributed to researchers and inoculant producers in the tropics. Improved techniques and systems for more dependable effective nodulation and utilization of legume-fixed nitrogen in cropping systems have been developed.

NifTAL researchers realized the benefits of collaborative efforts and have established international linkages through the International Network of Legume Inoculation Trials (INLIT). More than 200 LDC scientists from 53 nations have participated in the INLIT program.

Over 200 scientists have been educated through NifTAL's Legume/Rhizobium Technology training courses. Nine 6-week training courses have been held in Hawaii, Africa, Asia, and Latin America. Linkages have been established with the Bean and Cowpea and Peanut Collaborative Research Support Programs (CRSPs), with IRRI for rice, IITA for cropping systems in subhumid tropical areas of Africa, CIAT for common beans and legume forages, and IBPGR for collection, conservation and utilization of plant genetic resource materials.

A BNF Regional Resource Center has been established in Thailand and one is currently planned for Senegal for nitrogen-fixing trees. Hassan II University in Morocco has expressed interest in establishing a BNF Regional Resource Center in Morocco. University of Costa Rica and Zamorano University in Honduras have also expressed interest in establishing a center. The United Nations Development Programme (UNDP) has been working with NifTAL on N_2-fixing systems in lowland rice. The Government of Mexico has signed an agreement of intention to collaborate with the University of Hawaii in BNF programs.

A list of NifTAL network cooperators and officials familiar with NifTAL activities is attached.

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COMPLETE LISTING OF NifTAL NETWORK COOPERATORS PLUS OFFICIALS
FAMILIAR WITH NifTAL ACTIVITIES
(as of January 23, 1986)

* = NifTAL Network Cooperator
** = Official knowledgeable about NifTAL activities in country
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LISTING OF NifTAL NETWORK COOPERATORS PLUS OFFICIALS FAMILIAR WITH NifTAL PAGE 4

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LISTING OF NifTAL NETWORK COOPERATORS PLUS OFFICIALS FAMILIAR WITH NifTAL PAGE 5

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NEPAL

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NIGERIA

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PAKISTAN

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PEOPLES REPUBLIC OF CHINA

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* MRS. CHAI S.Y., YUNNAN PROVINCIAL ACD. OF AG. SCI., KUNMING, YUNNAN, PEOPLES REPUBLIC OF CHINA

LISTING OF NifTAL NETWORK COOPERATORS PLUS OFFICIALS FAMILIAR WITH NifTAL PAGE 6

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IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX H - KEY UNIVERSITY OF HAWAII PROJECT PERSONNEL

Revised:4/29/86: Revised 5/8/86
DIMPEX

Improved Biological Nitrogen Fixation thru Biotechnology

University of Hawaii - Key Project Personnel

B. Ben Bohlool, Soil Microbiologist. Received Ph.D. and M.S. from the University of Minnesota with emphasis in the areas of plant and soil microbiology. After two years of postdoctoral experience in microbial ecology at the University of Minnesota and the University of Wisconsin, he was appointed Research Scientist at the Cawthron Institute, New Zealand and spent three years there. He joined the faculty of the University of Hawaii in 1976 and is currently Professor of Soil Microbiology, Department of Agronomy, Soil Science. He has coordinated programs in the role of microbiology in agricultural development for third-world countries. His research work in microbiology has been diverse, ranging in scope from purely fundamental to highly applied. He has over 40 scientific publications dealing with soil and plant microbiology, biological nitrogen fixation, immunological techniques, thermophilic microbes and high-terperature systems, as well as aquatic and pollution microbiology. He was the recipient of the Alexander von Humboldt stiftung (Germany) in 1979 and spent 6 months with the molecular genetics group at the Max-Planck Institute in Köln, Germany. In January of 1985, he was appointed the Director of the Nitrogen Fixation for Tropical Agricultural Legumes (NifTAL) project.

Key Project Personnel

B.B. Bohlool

J.P. Roskoski. Microbial Ecologist. Ph.D. in Forest Ecology. Rockefeller Postdoctoral Fellowship to study nitrogen cycling in tropical agroecosystems in Mexico. Two years additional experience in Mexico studying fast-growing nitrogen fixing tree ecology, physiology, and management. Adjunct Assistant Professor, University of Arizona for two years. Head of Research section, Latin American network coordinator and associate director of the NifTAL Project from 1983. Research at NifTAL included nitrogen fixation by leguminous trees and in tropical cropping systems. Consulted for NAS/BOSTID CRG Program. Research interests include nitrogen fixation, leguminous trees, tropical cropping systems, nutrient cycling in natural and agro-ecosystems, tropical natural resource management.

P. Singleton. Agronomist. Ph.D. in Agronomy. During three years at NifTAL conducted research on the ecological and management factors that condition the response of legumes to inoculation with Rhizobium and host control of nodulation and nodule processes. Also African coordinator for INLIT experiments. Integrated several grants into the NifTAL core program.

C. Van Kessel. Plant Physiologist. Ph.D. in Biology/Crop Sciences. Prior to graduate work spent three years as a agricultural consultant in Cameroon, Africa, 2 years studying leguminous trees in Mexican coffee plantations, and 2 years investigating the physiology of nitrogen fixation by annual legumes. During two years as a research associate at NifTAL conducted research on physiology of nitrogen fixation by tropical legumes and field methods to quantify nitrogen fixation.

R. Davis. Soil Scientist. Following receipt of a Ph.D. in Soil Biology worked 22 years as a research scientist with the Agriculture Research Service of the USDA. During this time activities included research on nitrogen fixation, overseas assignment with USAID, and water quality work. Since 1979 has been Regional Coordinator for Asia at NifTAL, training director for two years, and since 1982 has been overall coordinator of the NifTAL Outreach Program.

P. Somasegaran. Microbiologist. After receipt of doctorate in 1977 joined NifTAL Project as chief microbiologist. Research activities included: strain selection, serology, and rhizobial ecology. Also foreign training instructor and technical director for NifTAL's international courses in Rhizobium technology. Curator for NifTAL's culture collection and consultant for various agencies on inoculant production facilities.

H. Hoben. Biochemist. Prior to joining the NIfTAL Project was engaged in toxicological studies on environmental pollutants, investgations on natural toxins in foods, and protein synthesis studies. In nine years at NifTAL has planned, coordinated, and taught BNF technology training courses, conducted research on Rhizobium ecology, inoculant production and serology, established an antiserum bank and helped develop small to medium size inoculant production fermenters.

Key Project Personnel (Continued)

Karean Zukeran. Project Manager. Associated with NifTAL's numerous activities on local, national, and international levels since 1976. Head the administrative Section with responsibility for daily operations of project, management of all aspects of personnel, procurement, and support activities including project organization, policy develeopment, national/international coordination and workshops. Affiliated with University of Hawaii since 1969.

Princess Ferguson. Information Specialist. Ten years experience in editorship, journalism, and print media. Has written a weekly column for local newspaper for five years. During last four of seven years at NifTAL has served as (1) 11/81 - Information Assistant and (2) 4/84 - Public Infomation Officer. Present duties include editing BNF BULLETIN newsletter, plus other books, pamphlets, and brochures produced by NifTAL. Supervises activities of communication section which include library, art, graphic, photographic, and audio-visual support.

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX I - UNIVERSITY OF HAWAII ADDITIONAL CONTRIBUTION TO THE PROJECT, BUT NOT CHARGED TO THE PROJECT

Revised:4/29/86: Revised 5/8/86
DIMPEX

ADDITIONAL CONTRIBUTIONS OF THE UNIVERSITY OF HAWAII

ITEM	REPLACEMENT COST/YEAR	TOTAL FOR 9 YEARS*
Laboratory Space (6000 sq. feet @ $3)	18,000	198,478
Office/Library Space (6000 sq. feet @ $3)	18,000	198,478
Experimental Fields (4 sites totaling 63 acres @ $300)	18,900	208,402
Equipment		
Scientific Instruments	226,289	226,289
Laboratory Glassware	100,000	100,000
Field Equipment	34,550	34,550
Office Equipment	54,099	54,099
Library Equipment	10,728	10,728
Facilities Maintenance Equipment	27,352	27,352
Vehicles	46,805	46,805
TOTAL	554,723	1,105,181

* Costs for rental of lab, office, and field space over 9 years are based on a 5% increase per year. Vehicles, scientific and non-scientific equipment, and glassware may not exist or have any value after 9 years.

University of Hawaii experts who can contribute time and Effort Towards Accomplishment of Objectives

Name	Degree	Position	Specialty and Expertise
ALLEN, R.	Ph.D.	Professor	Electron Microscopy and Protozoology
ALVAREZ, A.	Ph.D.	Associate Professor	Bacterial Plant Pathology and Monoclonal Antibodies
BARTHOLOMEW, D.	Ph.D.	Professor	Crop Physioloogy and Ecology
BENEDICT, A.	Ph.D.	Professor and Director of Monoclonal Antibody Ctr.	Immunology, Hybridoma Technology
BERGER, L.	Ph.D.	Professor	Industrial Microbiology and Biotech.
BOHLOOL, Ben	Ph.D.	Professor and Director NifTAL Project	Microbiology, Biotechnology and BNF
BREWBAKER, J.	Ph.D.	Professor and Director NFTA	Plant Genetics and BNF Trees
DEMB, A.	Ed.D.	Assistant Director of HITAHR	Administration and Social Policy
EL-SWAIFY, S.	Ph.D.	Professor and Dept. Chairman	Soil Errosion and Physics
HABTE, M.	Ph.D.	Associate Professor	Soil Microbiology, Mycorrhizae, and BNF
HUMPHREY, T.	Ph.D.	Professor	Molecular Biology of Differentiation, DNA Engineering and Biotechnology
IKAWA, H.	Ph.D.	Associate Professor	Soil Taxonomy, Networking
MANDEL, M.	Ph.D.	Professor	Molecular Genetics, DNA Engineering and Biotechnology
McARTHUR, H.	Ph.D.	Coordinator for International Program	Anthropology, International Development
MUELLER-DOMBOIS, D.	Ph.D.	Professor	Botany and Plant Ecology

Name	Degree	Position	Specialty and Expertise
PATIL, S.	Ph.D.	Professor and Director of Biotech Program	Plant Pathology and Biotechnology
ROBERT, F.	Ph.D.	Assistant Professor	Microbial Ecology and BNF
ROTAR, P.	Ph.D.	Professor	Agronomy and Pasture Management
SILVA, J.	Ph.D.	Professor	Agronomy and Biometrics
STILES, J.	Ph.D.	Associate Professor	Molecular Biology of Yeasts; Biotechnology
TANG, C.S.	Ph.D.	Professor	Plant Biochemistry and Allelopathy
UEHARA, G.	Ph.D.	Professor and Director of IBSNAT	Soil Physics and Crop Models
YAMAMOTO, H.	Ph.D.	Professor	Plant Biochemistry and Liposome Packaging

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 336-4177

APPENDIX J - Environmental Threshold Determination
Initial Environmental Examination

Revised:4/29/86: Revised 5/8/86
DIMPEX

mproved Biological Nitrogen Fixation Through Biotechnology

INITIAL ENVIRONMENTAL EXAMINATION

PROJECT LOCATION:	WORLDWIDE
PROJECT TITLE:	Improved Biological Nitrogen Fixation Through Biotechnology
PROJECT NUMBER:	936 4177
LIFE OF PROJECT:	Ten Years
IEE PREPARED BY:	Mary E. Mozynski
DATE:	April 21, 1986
ACTION RECOMMENDED:	NEGATIVE DETERMINATION

CONCURRENCE: _____

DATE: 5/30/86

Appendix J
April 21, 1986

MEMORANDUM TO: S&T/FA, Dr. Anson R. Bertrand

FROM: S&T/AGR, Tejpal Gill

SUBJECT: Environmental Threshold Determination

REF: Improved Biological Nitrogen Fixation Through Biotechnology - 936-4177

On the basis of the Environmental Threshold Determination, I recommend that you make the following determination:

__X__ 1. The proposed agency action is not a major Federal action which will have a significant effect on the human environment.

_____ 2. The proposed agency action is a major Federal action which will have a significant effect on the human environment, and:

_____a. An Environmental Assessment is required; or

_____b. An Environmental Impact Statement is required.

The cost of and schedule for this requirement is fully described in the referenced document.

_____ 3. Our environmental examination is not complete. We will submit the analysis no later than _____ with our recommendation for an environmental threshold decision.

Approved: Anson R. Bertrand

Disapproved _____

Date: 5/30/86

Clearance:Environmental Officer Carroll Collier
Carroll Collier

MMozynski 4/21/86

Improved Biological Nitrogen Fixation through Biotechnology

ENVIRONMENTAL ANALYSIS

I. Project Description

This project is designed to: 1) increase crop productivity; 2) reduce inefficient use of imported agricultural inputs; and 3) encourage sustainable agricultural production systems. It will promote the use of biological means to fix nitrogen in the soil to increase the availability of nutrients for plants. Its workplan is a multi-pronged design that involves research, training, information, networking, technical assistance and private enterprise. Products that emerge through research and development will be field tested under the auspices of the Regional Resource Centers. Implementation of the technology (production and marketing) will be facilitated through an incentive system by way of LDC small businesses.

Nitrogen and other essential nutrients removed from the soil must be replenished, in one way or other, to ensure sustained agricultural productivity. There are symbiotic microorganisms that can satisy the nitrogen needs of plants by directly incorporating into their proteins, nitrogen gas (N2) which is the major constituent of air. Successful use of biological nitrogen fixation can reduce the potential for nitrate contamination of ground water caused by the use of nitrogen fertilizers.

These symbiotic microorganisms generally will have little or no direct effect on the physical and natural environment as defined under Environmental Procedures 22, CFR Section 216.2 (a), (2), (i), (ii), (iii), (v), and (xiv). The University of Hawaii will assure that: 1) no research involving gene alterations and/or manipulation of already available altered genes of the microorganisms will be undertaken until the experimental protocols and laboratory facilities have been reviewed by appropriate Federal regulatory agencies and/or safety review boards; 2) appropriate U.S. Government and international quarantine regulations involving experimentation with and movement of rhizobium and plants potentially containing infectious plant diseases will be rigorously adhered to and in the event that the project leaders are not up-to-date of such regulations, they will contact the USDA's Animal and Plant Health Inspection Service (APHIS) for advice; and 3) if any pesticides are to be used in the project an environmental assessment will be prepared on such pesticides prior to their use under this project.

As research progresses, it is expected that new microorganisms will be developed. It is difficult to assess the impact of these microorganisms at this time since their exact nature is unknown. The field testing of such products after development will follow all appropriate Federal and International regulations. The anticipated impact of these products when ready for field testing will be evaluated on a case-by-case basis.

II. Recommendations

From the discussion above and the analysis contained on the following "Impact Identification and Evaluation Form," it is determined that the initial implementation of this project will not have a significant direct affect on the environment. A negative determination is recommended.

Improved Biological Nitrogen Fixation Through Biotechnology

ENVIRONMENTAL ANALYSIS

IMPACT IDENTIFICATION AND EVALUATION FORM

Impact Areas and Sub-Areas	Impact Identification *
A. LAND USE	
1. Changing the character of the land through:	
a. Increasing the population	N
b. Extracting natural resources	N
c. Land clearing	N
d. Changing soil character	H +
2. Altering natural defenses	N
3. Foreclosing important use	N
4. Jeopardizing man or his work	N
5. Other factors:	
Reducing pesticide pollution and contamination	N
B. WATER QUALITY	
1. Physical state of water	N
2. Chemical and biological states	L+
3. Ecological balance	N
4. Other factors	N
C. ATMOSPHERIC	
1. Air additives	N
2. Air pollution	N
3. Noise pollution	N
4. Other factors	N
D. NATURAL RESOURCES	
1. Diversion, altered use of water	N
2. Irreversible, inefficient commitments	N
3. Other factors	N

E. CULTURAL

1. Altering physical symbols	N
2. Dilution of cultural traditions	N
3. Other factors	N

F. SOCIO-ECONOMIC

1. Changes in economic/employment patterns	L +
2. Changes in population	N
3. Changes in cultural patterns	L +
4. Other factors	L +

G. HEALTH

1. Changing a natural environment	N
2. Eliminating an ecosystem element	N
3. Other factors	
Reducing pesticide poisoning	N
Reducing nitrates in water	L +

H. GENERAL

1. International impacts	L +
2. Controversial impact	U
3. Larger program impacts	L +
4. Other factors	N

I. OTHER POSSIBLE IMPACTS (not listed above)

1. Introduction of new plant species	U
2. Agricultural chemicals	L +
3. Other factors	N

* The following symbols are used for Impact Identification

N = No environmental impact
L = Little environmental impact
M = Moderate environmental impact
H = High environmental impact
U = Unknown environmental impact

+ = Beneficial impact
- = Negative impact

MMozynski:4/21/85

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX K - Publications of Collaborative Experiments with LDC Scientists

Revised:4/29/86: Revised 5/8/86
DIMPEX

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Results of collaborative experiments with NifTAL were reported in the following papers:

PATERNO, E.S. 1985. Utilization of Microbial Associations in Peanut Production: Rhizobium. Paper presented during the First National Peanut Consultation and Peanut-CRSP Review held at PCARRD Headquarters on February 7 - 8, 1985.

PATERNO, E.S. 1984. Rhizobium Technology in Crop Production. PCARRD Quarterly Seminar.

PATERNO, E.S. 1984. Legume Inoculation. Paper presented during the 1st Orientation Training/Seminar in Soybean Production Technology for Rice-Based Areas. PCARRD, Los Baños, Laguna. December 11 - 13, 1984.

PATERNO, E.S. 1983. Legume Inoculation: Problems and Prospects. Inaugural Professorial Lecture delivered at the Dept. of Soil Science Lecture Hall. July 13, 1983.

PATERNO, E.S. 1983. Status and Needs of Research on Legume Varietal Development and Cultural Management. Paper presented during the Conference on Status and Needs of Legume Research, Extension and Instruction at UPLB held on September 27, 1983 at the Plant Pathology Auditorium, UPLB.

PATERNO, E.S., N.T. ARMONES, V.Z. PERDIDO, B.G. MAGTAGNOB and M.L.Q. SISON. 1983. Legume Inoculation with Rhizobia: Field Trials. Terminal Report. PCARRD Project.

PATERNO, E.S., N.T. ARMONES, V.Z. PERDIDO and B.G. MAGTAGNOB. 1982. Legume Inoculation with Rhizobia: Field Trials. Annual Report. PCARRD Project.

Additional Comment:

Results of the INLIT program have increased awareness on the benefits of legume inoculation among policy makers, researchers and extension workers.

LIST OF PUBLICATIONS

1. Shahjahan, M., D. Begum and M.S. Islam, 1981. Biological Nitrogen Fixation by Grain Legumes. Proceedings of the National Workshop on Pulses, BARI, Joydebpur, P. 226-231.

2. Shahjahan, M. and Rafiqul Islam, 1984. Preliminary Screening of Rhizobia strains for Mungbean. Bangladesh J. Agri. Res. 9(2): 130-135 (1984).

3. Shahjahan, M. Response of Chickpea to Rhizobium inoculation. Accepted to publish in the Bangladesh Journal of Agricultural Research.

4. Shahjahan, M. 1981. Response of Chickpea to Rhizobium inoculation and N-fertilization. BARI Annual Report, 1980-81, Joydebpur, Dhaka, P-59.

5. Shahjahan, M. 1981. Response of Chickpea to Rhizobium inoculation and P-fertilization, BARI Annual Report, 1980-81, Joydebpur, Dhaka, P-60.

6. Shahjahan, M. 1982. Preliminary Screening of strains of Rhizobia for mungbean. BARI Annual Report, 1981-82, Joydebpur, Dhaka, P-66-67.

7. Shahjahan, M. 1982. Response of mungbean to Rhizobium inoculation. BARI, Annual Report, 1981-82, Joydebpur, Dhaka, P. 67.

8. Shahjahan, M. 1983. Response of chickpea to Rhizobium inoculation (NifTAL trials). BARI Annual Report, 1982-83, Joydebpur, Dhaka, P. 53-54.

9. Shahjahan, M. 1983. Response of mungbean to Rhizobium inoculation. BARI Annual Report, 1982-83, Joydebpur, Dhaka, P. 59-60.

10. Shahjahan, M. 1983. Response of mungbean to Rhizobium inoculation and N-fertilization, BARI Annual Report, 1982-83, Joydebpur, Dhaka, P. 60.

List of research publications of A.R.M. Solaiman

1. Effectiveness of locally prepared and imported inoculants of soybean in Bangladesh M.S. Hoque, A.R.M. Solaiman and Z.H. Bhuiya. Thai. J. Agric. Sci. 15:127-134. 1982.

2. Nitrogen Fixation work on soybean at Bangladesh Agril. University. M.S. Hoque AB.M.S. Islam, A.R.M. Solaiman and Z.H. Bhuiya, Proc. National Workshop on Pulses. August, 1981.

3. Importance of Biological Nitrogen Fixation studies in Bangladesh. A.K.M. Habibullah, A.K. Poddar and A.R.M. Solaiman ADAB NEWS, Sept-Oct., 9(5),1982.

4. Performance of some localRhizobium inoculants and Nitrogin on nodulation, dry matter production and grain yield of Davis soybean, M.S. Hoque, A.R.M. Solaiman and Z.H. Bhuiya. ~~Bang.~~ Bangladesh J. Agric. Sci. 19(1),1983.

5. Effects of some locally prepared Rhizobium inoculants on nodulation and dry matter yield of soybean. A.R.M. Solaiman and M.S. Hoque, ~~Bang.~~ Bangladesh J. Agric. 8(1-4),1983.

6. Importance of Biological Nitrogen Fixation by non-legumes and freeliving organisms in Bangladesh. A.K. Poddar, A.K.M. Habibullah and A.R.M. Solaiman ADAB NEWS, March-April,1983 10(2).

7. Effects of different Rhizobium inoculants on nodulation, and N nutrition ~~[illegible]~~ in Soybean. A.R.M. Solaiman and M.S. Hoque. Proc. Int. Symp. on Soil Test Crop Response Correlation Studies. ~~(In Press).~~ 1984.

8. Field responses of groundnut (Arachis hypogea L.) to Rhizobium inoculant compared to urea nitrogen at farm and maximal fertility levels I. Nodulation, dry matter production, N content and N uptake. A.R.M. Solaiman and A.K.M. Habibullah. Bangladesh J. Agric. (Sent for publication).

9. Field responses of groundnut (Arachis hypogaea L.) to Rhizobium inoculant compared to urea nitrogen at farm and maximal fertility levels II. Yield and yield contributing characters. A.R.M. Solaiman and A.K.M. Habibullah. Bangladesh J. Microbiology (~~Sent~~ Accepted for publication).

Publications resulted from experiences at NifTAL

1. A.B.Guevarra, Y.Kitamura, A.S.Whitney and K.G.Cassman : A low-cost system for circulating nutrient solutions in pot studies. Crop Science Vol. 20, 110 - 112.(1980)

2. Y.Kitamura, A.S.Whitney and A.B.Guevarra : Legume nitrogen fixation as affected by plant competition for light and for soil nitrogen. Agronomy Journal. Vol.73, 395 - 398 (1981)

3. Y.Kitamura and J.Abe : Efficiency of N_2-fixation by tropical pasture legumes. Journal of Japanese Society of Grassland Science Vol. 28, 154 - 160 (1982).

Yukio Kitamura
National Grasslands Research Institute

Publications

- Influence of Molybdenum on Nitrogen Fixation by white clover in the Bogotá Savanah. Biological Nitrogen Fixation for Tropical Agriculture. Peter H. Graham, Susan Harris Editors, 161-165 , 1981

- Efecto de la interacción Nitrógeno por Molibdeno sobre la fijación por trébol blanco en suelo serie Techo. Suelos Ecuatoriales 12(1) 177-176, 1982

Efecto de la inoculación fertilización y encalado sobre la fijación simbiótica de Nitrógeno en cultivos de arveja y trébol en suelos de la serie Tibaitatá. Suelos Ecuatoriales 12(1) 177-181, 1982.

- Efecto del Fósforo en la respuesta simbiótica del trébol blanco en diferentes fases de su desarrollo. Suelos Ecuatoriales 1984.

IMPROVED BNF THROUGH BIOTECH

PROJECT NO. 936-4177

APPENDIX L - Summary Responses to Questionnaire on the Value of NifTAL Training

Revised:4/29/86: Revised 5/8/86
DIMPEX

March 1986

QUESTIONNAIRES

In late 1985, a questionnaire was developed to be sent to participants of NifTAL training courses and others receiving the BNF BULLETIN Newsletter. The attached questionnaire responses have been divided into three categories.

The first category includes responses from former training course participants. The second and third categories include responses from non-participants in developing and developed countries--these respondents answered a slightly modified version of the questionnaire. Distribution of the questionnaire was made to approximately 1500 persons and to date more than 60 completed responses have been returned.

Each of the attached three response categories include a narrative analysis of relevant responses, a list of respondent's names, a report including each question with corresponding answers received, and any additional comments made by respondents.

NARRATIVE ANALYSIS

This questionnaire was directed to approximately 350 persons who had been identified as having been involved in NifTAL training. Twenty-five former participants responded to this questionnaire. These respondents represent a variety of NifTAL BNF Technology courses (1977 to 1984) as well as interns and visiting scientists.

All respondents reported gains in scientific skills and techniques, understanding, and even in supervisory and managerial skills. 50% had changed positions as a result of training. Most of these reported being deeply involved in BNF research or promotion programs. In fact, 95% presently work in BNF. Over 50% are research scientist and the remainder are either university teachers or inoculant producers. One-third reported that their experience had resulted in a publication. Twenty-one of the 25 respondents have used the NifTAL training manual to train others.

Nearly 50% have sought and received funding for research related to BNF from several different organizations. Nearly all had received cultures, inoculant, antisera, seeds, or reprints and had used NifTAL Rhizobium in their research work both in lab and field studies. These studies represent a wide variety of experiements with many different legumes. NifTAL strains gave satisfactory results and these strains have been recommended to in-country inoculant producers by over 50% of the respondents. Nearly all respondents make inoculant for research purposes while about 40% make inoculant for commercial purposes and nearly 50% of the respondents have helped set up an inoculant facility in their country. One-half of the respondents have become collaborators in the INLIT trials.

In response to a request to rank NifTAL's impact on BNF in their country, 55% ranked NifTAL at 8 or above (on a scale with 10 highest). Thirteen respondents knew of no other organization working on BNF in their country besides NifTAL.

RESPONDENTS

Name	Country	Comments
Abdel El Shakie	Sudan	
Ahmed A. A. Mahdi	Sudan	
Garg	India	
P. Thirukkumaran	Sri Lanka	
E. Paterno	Philippines	Results of the INLIT program have increased awareness on the benefits of legume inoculation among policy makers, researchers, and extension workers
Gayaga Ocampo	Philippines	
Jbel Espinosa de M	Mexico	
Fe G. Torres	Philippines	
E. Cuautle-Fabian	Mexico	
N. D. Purwantari	Indonesia	
Dilwara Begum	Bangladesh	
Raul Varela G	Brazil	
Amanda L. de Yunda	Colombia	
A.R.M. Solaiman	Bangladesh	
Vincent Michelina	Venezuela	
Saninjit Singh	Trinidad, W.I.	
Visitacion Perdido	Philippines	
Chirasak Arunsri	Thailand	
Marcio Voss	Brazil	
Md Shahjahan	India	
Wichian Magtoon	Thailand	
Hussein Kamel	Egypt	
Ronald F-Cerrato	Mexico	
Messaud Lahbib	Senegal or Mali	
Anabal Alvarez	Uruguay	

March 1986

QUESTIONNAIRE RESPONSES

Trainee Directed - Developing Countries

1. Were you a participant in a NifTAL training course?

25 - yes

4 - 83/ICRISAT; 2 - 77/Maui; 2 - 80/Porto Alegre; 2 82/Maui; 1 each: 84/NifTAL, Maui; NifTAL, Maui; 81/Maui; 76/Maui; 81/Chapingo, Mexico; 82/Thailand; 79/Porto Alegre; 81/Venezuela; 80/MARDI

2. Did this training enhance your ability to perform scientific work?

25 - yes

2A In what way did this training make a difference in your work?

A new line of research has been initiated; better understanding; in producing better quality inoculants; trained microbiologists for further study; it has enhanced my supervisory ability on BNF by training others along this line and preparing research proposal; acquired skills of handling Rhizobium in the lab, greenhouse, and field procedures. I have now done research in Veracruz and have a project; it strengthened my capability to do research on BNF-improved skills; better methods of transferring and fermenter technology; it has increased my knowledge and skill to manage research on legume/Rhizobium symbiosis; isolation of Rhizobium strains, inoculum production; my knowledge of BNF were improved. I was able to do research in a more efficient way. Also, I got new ideas from this training; the training offered me enough practical knowledge in the field of legume/Rhizobium technology; now I handle better BNF than before; establishment of Rhizobium inoculant facility; it increased my knowledge on the importance of Rhizobium inoculant for leguminous crops; Rhizobium inoculant production; introduced a new technique in preparation of Rhizobium inoculant; by allowing the opening of BNF research line in our institute; I was able to supervise my section regarding Rhizobium/legume symbiosis much better because I had a better understanding of the subject matter both theoretically and practically after my training.

3. As a result of the training you received from NifTAL, has your position changed?

13 - yes; 11 - no; 1 - no response

3A Please specify. Answers (11): Head of BNF Department; research officer in charge of microbiology unit; quality control officer on inoculants produced by the Bureau; yes - production of inoculants for beans; I lost my job because my boss saw me as a strong competitor; I am looking after inoculant production independently now; work individually on BNF; I have been working as an Associate Investigator of the national BNF Project/Bangladesh after getting such training; I

am head of a BNF project; inoculant production; promoted to Senior Scientific Officer and coordinating BNF work; actually there are 7 different projects on BNF in our section; after my training in 1977, Soil Biologist, Soil Research Division, Bureau of Soils

4. Have you used the NifTAL training manual to train others?

21 - yes; 4 - no

4A&B 6 - no response. Answers: 1983-85: Sudan/students; Since 1983: Sudan; Past 4 years: in the lab, Sri Lanka Soybean Project; 1978-present: Bureau of Soils; 1981 to date: Veracruz, Mexico; 1980-81: Postgraduados, Chapingo, Mexico; 1984: here in Palmira; Oct 1984: National University/Bogota; At my institute informally since 1982: Venezuela; 1983: Trinidad; every year in Thailand; 1982-85: Londrinas, Brazil; 1984: in the field of my University; after come back in my country at National Research Center; May-Jun 1980: NifTAL/Colegio Postgraduados Course

5. Are you working in BNF? In what capacity?

24 - yes; 1 - no response

5A In what capacity: 16 research scientists, 8 university teachers, 6 inoculant producers; Training & Transfer Technology Officer

6. Were you a visiting scientist at NifTAL?

2 - yes; 23 - no

7. Were you a graduate student supported by NifTAL?

2 - yes; 22 - no; 1 - no response

8A If you were a visiting scientist, who did you work most with at NifTAL?

23 - no response; 2 answers: "Somasegaran," "Somasegaran & Hoben"

8B For how long?

2 responses: 2 months; 5 months

9. Did your experience result in a publication?

9 - yes; 8 - no; 1 - NA; 7 - no response

9A Note citation. See lists attached.

10. Have you sought and received funding for research related to BNF?

12 - yes; 13 - no

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10A From whom?

UNEP/Ag Fund & Sudan Government; UNEP; USAID (Pending); CRSP/PCAR; Florida State University; BARC, Dhaka; Univ Oriente, Maturia, Venezuela; UNDP; NifTAL PCARRD; Brazilian Government; BARC (submitted for approval); CONACY/Mexico; IFS (Sweden)

11. Have you received NifTAL Rhizobium cultures, inoculant, antisera, seeds, or scientific reprints?

22 - yes; 2 - no; 1 - no response

11A 12 - cultures; 11 - reprints; 6 - inoculant; 2 - seeds; 3 antisera; 2 - INLIT strains; 1 - all

12. Have you used NifTAL Rhizobium in your research work?

22 - yes; 2 - no; 1 - no response

12A Lab studies?

18 - yes; 1 - no; 6 - no response

12B Field studies?

18 - yes; 7 - no response

12C Please specify what and how:

Many strains for P, Cicer, Glycine, Acacia, Clitoria, Arachis in inoculant experimental material or as references TAL 1127 & 1132; selection of strains for local conditions; NifTAL strains were field tested and screened for compatability with local legume cultivars using Leonard jars; lab studies by comparing the NifTAL Rhizobium cultures with indigenous Rhizobium; bean and soybean inoculation trials in field to show response of nitrogen fixation. Good results with soybean and promising results with beans; INLIT; in soybean trials and field demonstrations, results good; cultivars of Lens culinaris, V. radiata, V. mungo, peat pased inoculum in field soybean strains; V. spp.; I have seen comparative effects of local vs. exotic strains. Local strains found as effective as the exotic (NifTAL) ones; Rhizobium inoculant of peanut, soybean, & mung to evaluate the performance of legume inoculants in the field; cowpea Rhizobium for numbers by test of yield trial; in preparing more than 4,000 inocula (one/feddu) yearly; by comparing NifTAL strains with Rhizobium strains isolated from Mexico; as comparative study with indigenous isolated in Philippines.

13. Have you received satisfactory results using NifTAL strains?

16 - yes; 2 - no; 7 - no response

14. Have you recommended our strains to inoculant producers in your country?

13 - yes ; 2 - no; 10 - no response Comments: yes "in some cases," & no "there isn't inoculant producers"

15. Do you make inoculant for research purposes?

21- yes; 4 - no

16. Do you make inoculant for commercial purposes?

9 - yes; 9 - no; 7 - no response Comments: yes "in future" & "hoping to make"

17 Have you helped set up an inoculant facility in your country?

12 - yes; 8 - no; 5 - no response

17A-D Where? How much inoculant is made in your country? For which legumes are inoculants being made in your country? How much inoculant is imported?

Khartoum, none, --, none
Sudan, experimental amounts for broad bean/chickpea/cowpea, little
--, no idea, chickpea/pigeonpea/mung/groundnut/soybean/Berseem/lucerne/-pea/Lentil, --
Sri Lanka, 20,000 1-acre packets, soybeans, 10,000
UP at Los Banos, 30 Kg/yr at WPLB, mungbean/peanut/cowpea/Leucaena, none
--, --, soybean/Leucaena, --
Bureau of Soils, 2-3 tons/yr, Mungo/peanut/soybean/beans/kudzo/reto, none
--, 200 T, soybean (90 or more %), none
AASAM, approximately 10-15M packets, all legumes, nil
at our lab, as much as required, L. culinaris/V. mungo/V. radiata/C. arietinum, only as required for research
--, there isn't, --, all
--, not commercially produced, soybean/groundnut/lentil/chickpea, inoculant is not sold commercially
In Venezuela, I don't know, soybean/black bean/Stylosanthes, I don't know
St. Augustine, yes, small scale, --
Bangkok, approximately 35 tons/year, soybean/peanut/mungbean, --
Brazil, + 900,000/200g., soybean/pisum, zero
Dhaka, --, one Dr. Idris is going to set up an inocualnt factory in Dhaka and I am giving there advisory services, --
In NRC, 25%, faba/lentil/groundnut/lupin/Berseem/etc., little-only for research
--, --, bean/alfalfa/faba/soy, none
different regions, 2-3 ton/year, mung/peanut/soybean/etc., none

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18. Have you participated in the INLIT program?

    11 - yes; 14 - no

18A When? 79-82, 81 to date; Since 82; 2 - 82-83; 80-81; 81; 82; 84; 80

18B What trials (A, B, C)? 10 -A; 1 - B

18C Which legume? 7 - soybean; 2 - peanut; 2 - *P. vulgaris*; 2 - mungbean; 1 - *Lens culinaris*; 1 - *V. radiata*; 1 - *Arachis hypogaea L.*

19. Please rate NifTAL's impact on BNF in your country. Use a scale of 1 (lowest) to 10 (highest).

4 - 10	1 - 6
1 - 9	2 - 5
1 - 8-9	2 - 3
7 - 8	2 - 2 (much has to be done)
2 - 7	1 - yes and 2 - no response

20. What other international research organizations are supporting BNF activities in your country?

    7 - no response
    Answers: UNEP/Unesco; UNEP; Institute of Fundamental Studies; USAID/BOSTID/ACIAR/JICA; none Vienna N-15 Project; CRSP; IOAE; Not Known; FAO; CIAT; ICRISAT; I don't know; BNFRC (NifTAL); USAID; ICARDA, USA Academy of Science (IAEA-15-N) (FAO-Leucanea); Unesco-MIRCEN (W. Africa, Senegal); none