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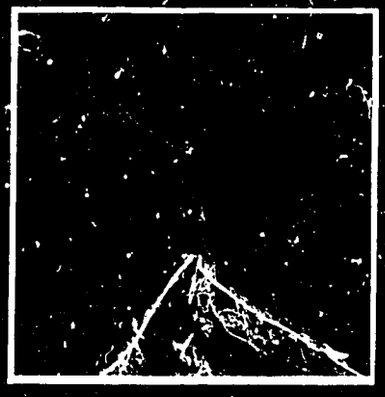

On the cover . . .

NifTAL

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1. Senior legume scientists learn advanced techniques in NifTAL's intern program.

2. Legumes can be self-sufficient for nitrogen thanks to the rhizobia housed in their root-nodules.

3. A Honduran researcher is trained in antisera production at a NifTAL course in Mexico.

4. International scientists receiving NifTAL training on *Rhizobium* strain selection in Malaysia.

5. Inoculated bean seeds on the right have the edge—10,000 rhizobia/seed.

6. One of over 200 scientists participating in NifTAL's International Network of Legume Inoculation Trials.

7. A refresher course on Inoculant Production and Quality Control taught at Hyderabad, India.

8. Common beans *(Phaseolus vulgaris)* respond markedly to inoculation with acid tolerant rhizobia in an Ultisol.

9. African researchers learn how to test for successful inoculant strains at NifTAL's Nairobi course.

NITROGEN FIXATION BY TROPICAL AGRICULTURAL LEGUMES

NFTAL
University of Hawaii/USAID

ANNUAL REPORT 1983

USAID Contract No. DAN-0613-C-00-2064-00
01/01/83-12/31/83

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UNIVERSITY OF HAWAII

*An Institute integrating the former Hawaii Agricultural Experiment Station and the Hawaii Cooperative Extension Service.

STATISTICAL SUMMARY

TITLE	: Nitrogen Fixation by Tropical Agricultural Legumes (NifTAL Project)
CONTRACT NO.	: DAN-0613-C-00-2064-00 (was AID/ta-C-1207)
PROJECT NO.	: 931-0613
PRINCIPAL INVESTIGATOR	: Dr. Jake Halliday
ADDRESS	: NifTAL Project and MIRCEN P.O. Box 0 Paia, Hawaii 96779, U.S.A.
CONTRACTOR	: University of Hawaii Department of Agronomy and Soil Science College of Tropical Agriculture and Human Resources 3050 Maile Way, Honolulu, Hawaii 96822.
CONTRACT PERIOD	: 06/16/82-12/31/86
REPORTING PERIOD	: 01/01/83-12/31/83
USAID FUNDS TO DATE	: $1,650,000
CURRENT YEAR EXPENDITURES	: $1,000,000
ESTIMATED 1984 FUNDS	: $1,350,000
TOTAL CONTRACT FUNDS	: $4,875,000
PROJECT OFFICE	: S&T/AGR/RNR
PROJECT MONITOR	: Dr. Lloyd R. Frederick

J Halliday

Submitted by
Principal Investigator

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In 1975, the United States Agency for International Development (USAID) contracted with the University of Hawaii to implement a project to help farmers in the developing countries step up production of high protein foods with reduced dependence on costly nitrogen fertilizers. This is being accomplished by and used to the fullest extent possible in tropical cropping systems.

The initial USAID contract drew on expertise in the College of Tropical Agriculture and Human Resources for establishment of an interdisciplinary unit called NifTAL*, dedicated to the application of technologies based on biological nitrogen fixation (BNF) to international development goals.

NifTAL is dedicated to the application of technologies based on biological nitrogen fixation to international development goals.

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 multitier training.

In 1979, NifTAL undertook to facilitate global, regional, and in-country networks to enable developing country scientists to gather multilocation data needed to answer key research questions. These networks have been effective in providing developing countries with access not only to the resource represented in NifTAL, but also to expertise of leading BNF scientists throughout the U.S., Europe, and Australia.

And because BNF scientists in neighboring countries recognized advantages in sharing their perspectives on how to apply their acquired expertise to crop production constraints, tighter networks have now emerged.

*NifTAL is an acronym from Nitrogen Fixation by Tropical Agricultural Legumes.

Recently, in-country networks have been forming to put in place the infrastructure needed to support the use of BNF technologies by farmers. These networks typically involve several government departments, non-governmental organizations, private entrepreneurs, and aid agencies.

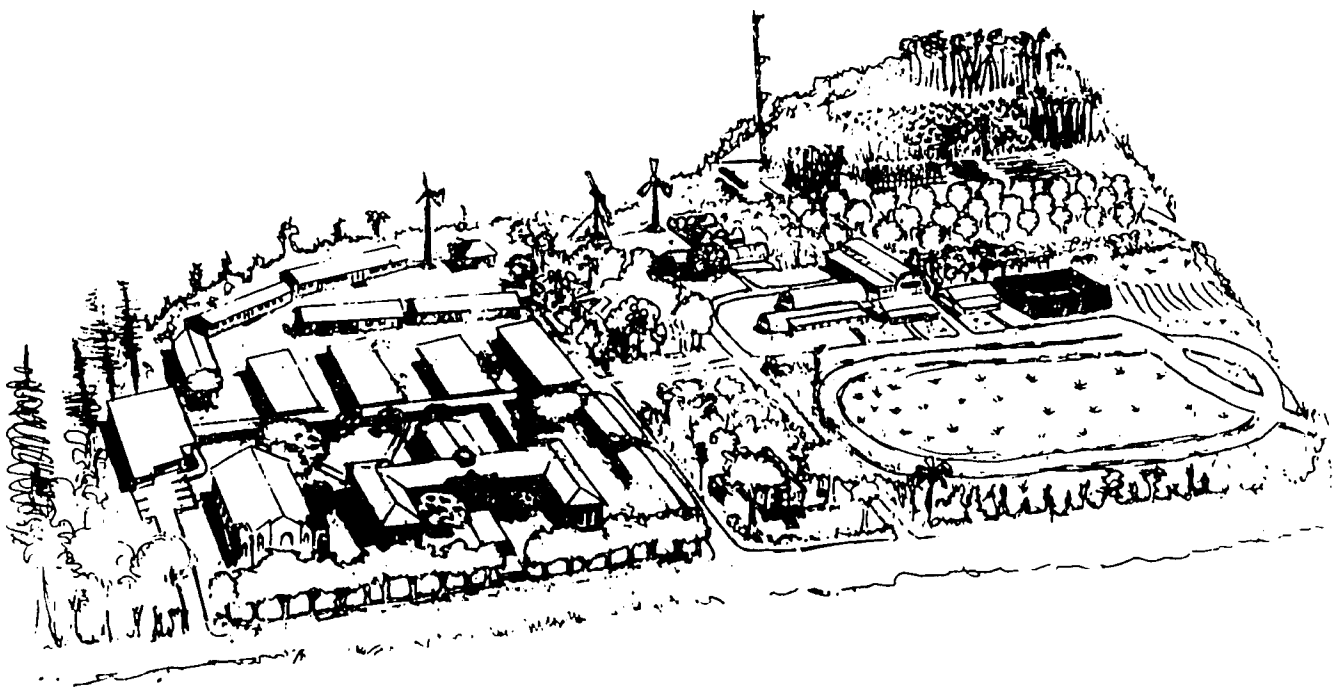
It has been a privilege for NifTAL to participate in this progression towards fuller use of biologically fixed nitrogen in tropical cropping systems. In meeting the challenge to tackle remaining constraints, we rely increasingly heavily on collaborative linkages with colleagues in the developing countries and elsewhere within the framework of a global BNF network.

The University of Hawaii acknowledges the continuing support by the USAID for NifTAL, initially through Contract AID/ta-C-1207 (1975–1982) and currently under DAN 0613-C-00-2064-00. NifTAL has grown in size and capability over the years. The USAID core budget for the NifTAL Project was the catalyst permitting establishment of an integrated, multidisciplinary program now augmented by many other awards from public, private, and international sources.

The University also recognizes the strong support of the Mayor of Maui and the Maui County Council for provision of the 25 acre NifTAL research campus.

Thanks are also due to the individual farmers and agribusinesses on Maui who provide land for experiments and host NifTAL's overseas visitors.

The USAID core budget for the NifTAL Project was the catalyst permitting establishment of an integrated, multidisciplinary program now augmented by many other awards from public, private, and international sources.



What a year for NifTAL! We have been tackling a new contract with a wider scope of work, adjusting to shifts of emphasis in research and training, and establishing a BNF Resource Center for South and Southeast Asia at Bangkok.

Wider scope of work. NifTAL's goal remains unchanged: to enable farmers in developing countries to increase production of high protein crops with reduced dependence on nitrogen fertilizer. NifTAL addresses this challenge through programs to maximize the benefits of biological nitrogen fixation (BNF) in tropical cropping systems. At one time NifTAL dealt exclusively with tropical grain legumes which have a direct role in human nutrition. Later, the tropical forage legumes were included. Given the many important non-food uses of legumes, especially the tree species, NifTAL is now mandated to address BNF aspects of all legumes with a role in development. 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. Thus NifTAL balances and complements the legume programs of the International Agricultural Research Centers and Collaborative Research Support Programs which focus on particular commodities and defined geographic areas.

Shift in research emphasis. In the past, NifTAL has concentrated on selection of superior strains of *Rhizobium* for use in inoculants for tropical legumes in harsh soil conditions. Although further advances with this approach can still be expected, satisfactory strains have been assembled for many legumes and for most soil conditions. Now it is time to develop an improved understanding of how to manage tropical cropping systems to optimize use of nitrogen fixed biologically.

New training goals. Since 1975, we have trained many key researchers through our postgraduate programs and short courses. In some countries, the technology is now in demand by farmers and by industrialists interested in legume inoculants. Thus NifTAL in now working with extension leaders on design of materials to explain the use of the technology to farmers. We are also delivering training on commercial scale production of quality inoculants.

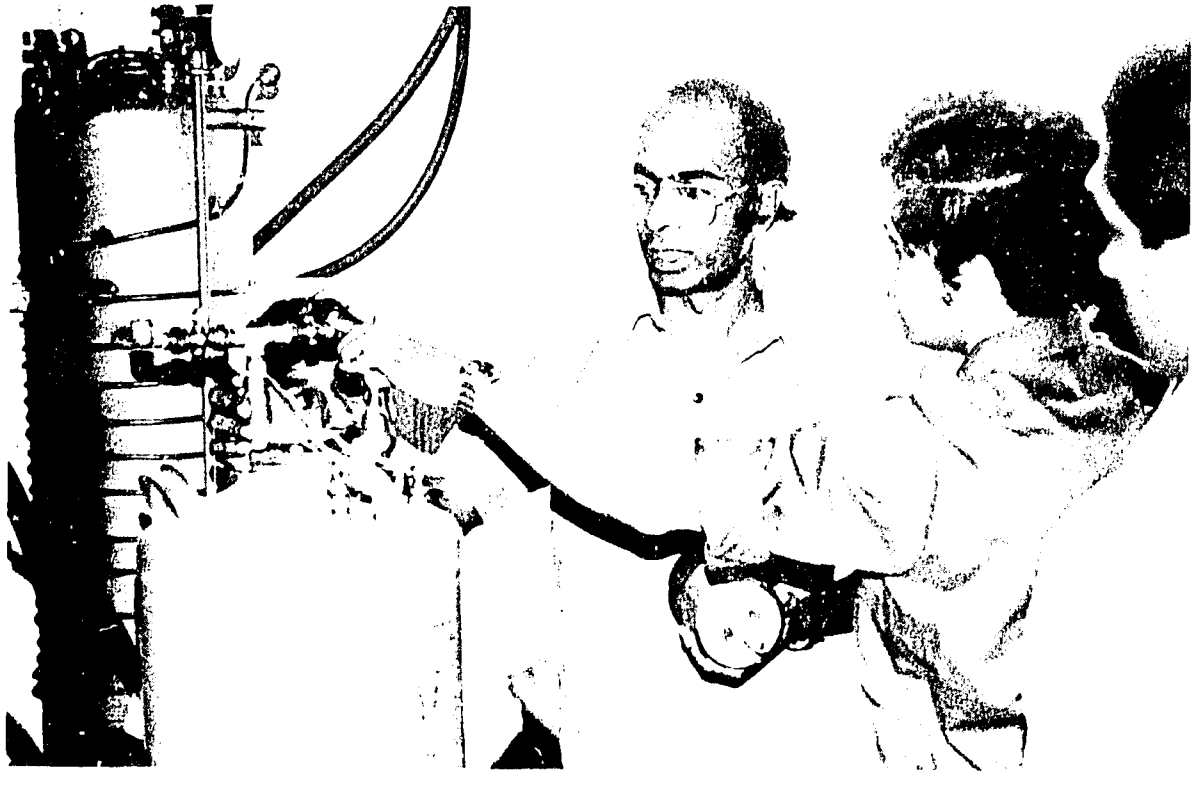
NifTAL is using microcomputer communication to further international development goals.

BNF Resource Center. National agricultural organizations of the developing countries have research programs on leguminous crops. Even when the approach is multidisciplinary, few legume teams can include professional *Rhizobium* specialists. A philosophy implicit in the NifTAL concept is assembly of a relatively large resource unit staffed by BNF specialists with a commitment to international development and equipped to service the needs of national legume programs. It is becoming increasingly difficult and costly to meet requests for specific support to national programs from our Hawaii base. NifTAL is taking an initiative to offer research support, technical and material services, and training oriented specifically at production constraints of the cropping systems of the South and Southeast Asian region. We are indebted to the Thailand Department of Agriculture for hosting the BNF Resource Center and for providing superlative facilities. Through this presence in the region, we expect to capitalize even further on the network approach to accomplishing research and facilitating the interchange of technical information.

1983

These were the themes that dominated this reporting year at NifTAL, and within this framework we are pleased to be recording real progress and positive achievement—read on!

(Above) Since 1975, NifTAL has trained many key researchers through our post-graduate programs and short courses. (Below) NifTAL is staffed by BNF specialists with a commitment to international development and is equipped to service the needs of national legume programs.



For those people in the huts and villages of half the globe, struggling to break the bonds of mass misery, we pledge our best efforts to help them help themselves for whatever period is required:

— not because the Russians may be doing it;

— nor because we seek their votes;

— but because it is right.

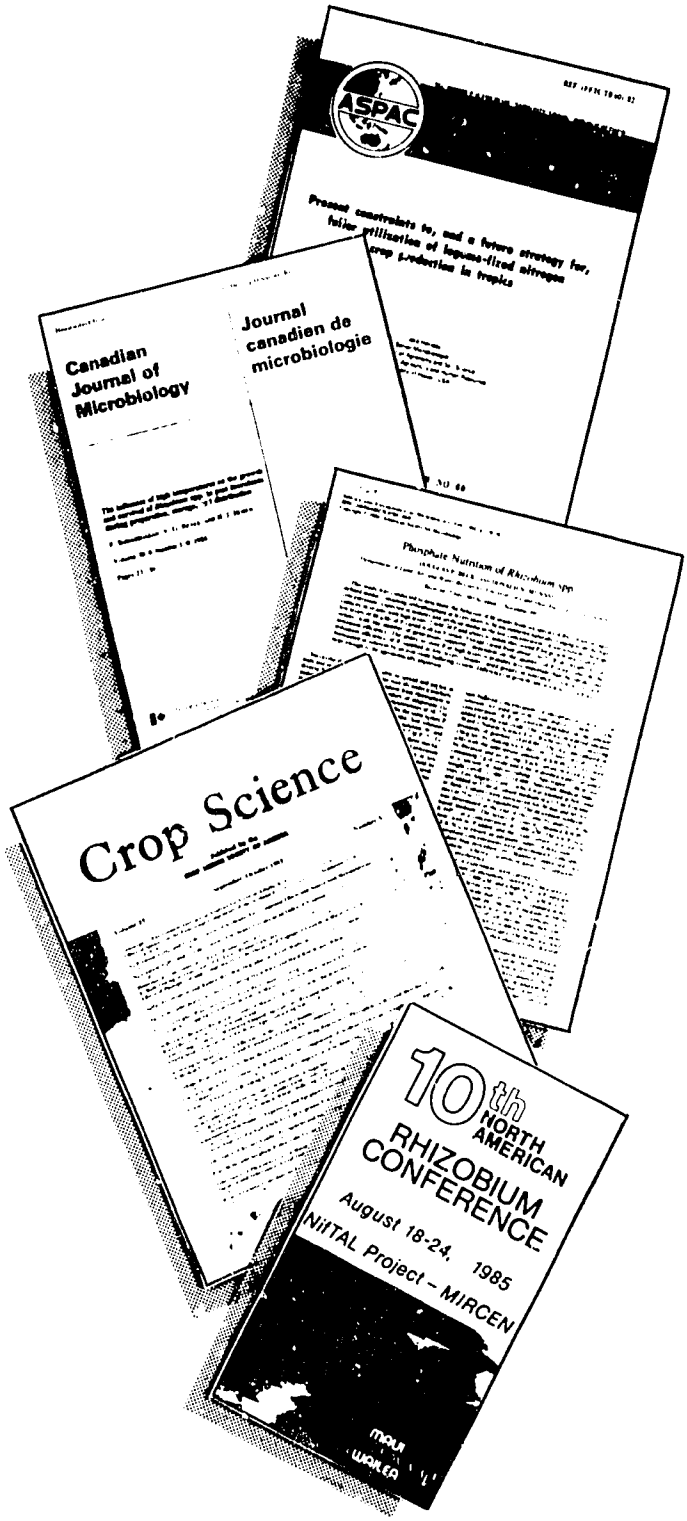
John F. Kennedy

The focus of the NifTAL Project is a process (biological nitrogen fixation) rather than a commodity or a discipline. In fact, NifTAL rallies the expertise of many disciplines to address a process which benefits an array of commodities.

Some constraints on fuller utilization of biological nitrogen fixation are technical and are being addressed through research. Economic and social factors also limit this technology as does lack of personnel trained in the skills required for research, development, and use of inoculants.

So, despite a seemingly narrow focus on biological nitrogen fixation, a wide range of activities is undertaken by NifTAL to meet our goal of ensuring that the process is used to the maximum extent possible in tropical cropping systems.

The sections which follow summarize progress to date, review specific accomplishments this year, and touch on future plans for each category of activity. A full workplan for 1984 is given in Appendix III. Readers interested in detailed research results are referred to the documents in Appendices I and II, each of which is available on request from NifTAL.



It is NifTAL's policy to disseminate research results through publication in appropriate journals and through conference and workshop presentations. In this way, data derived with contract support are made a part of the permanent scientific record, freely accessible for future reference.

Activities conducted by NifTAL:

- Regionalization of BNF support
- Training and communication
- Country and interagency projects
- International network trials
- Legume inoculant development
- Rhizobial germplasm development
- Rhizobial antisera services
- N management in cropping systems
- Soil stress tolerance research
- Economic analysis of BNF

Objective:

— to tailor development support services offered globally from NifTAL, Hawaii, to specific needs of countries of South and Southeast Asia.

Background

Many general 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 strategic use of legumes in tropical cropping systems, it has become necessary to tailor NifTAL outputs to the vastly differing cropping 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 towards that goal.

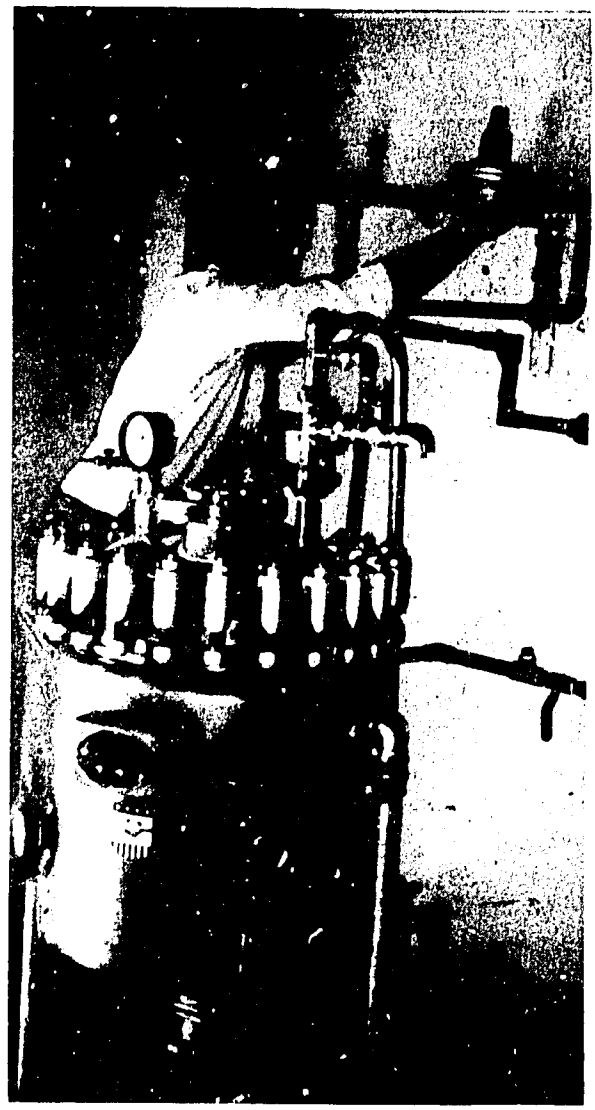
This step also makes it easier for USAID's Regional Bureaus and Country Missions to tap NifTAL resources in support of regional and country programs involving legume crops.

Accomplishments

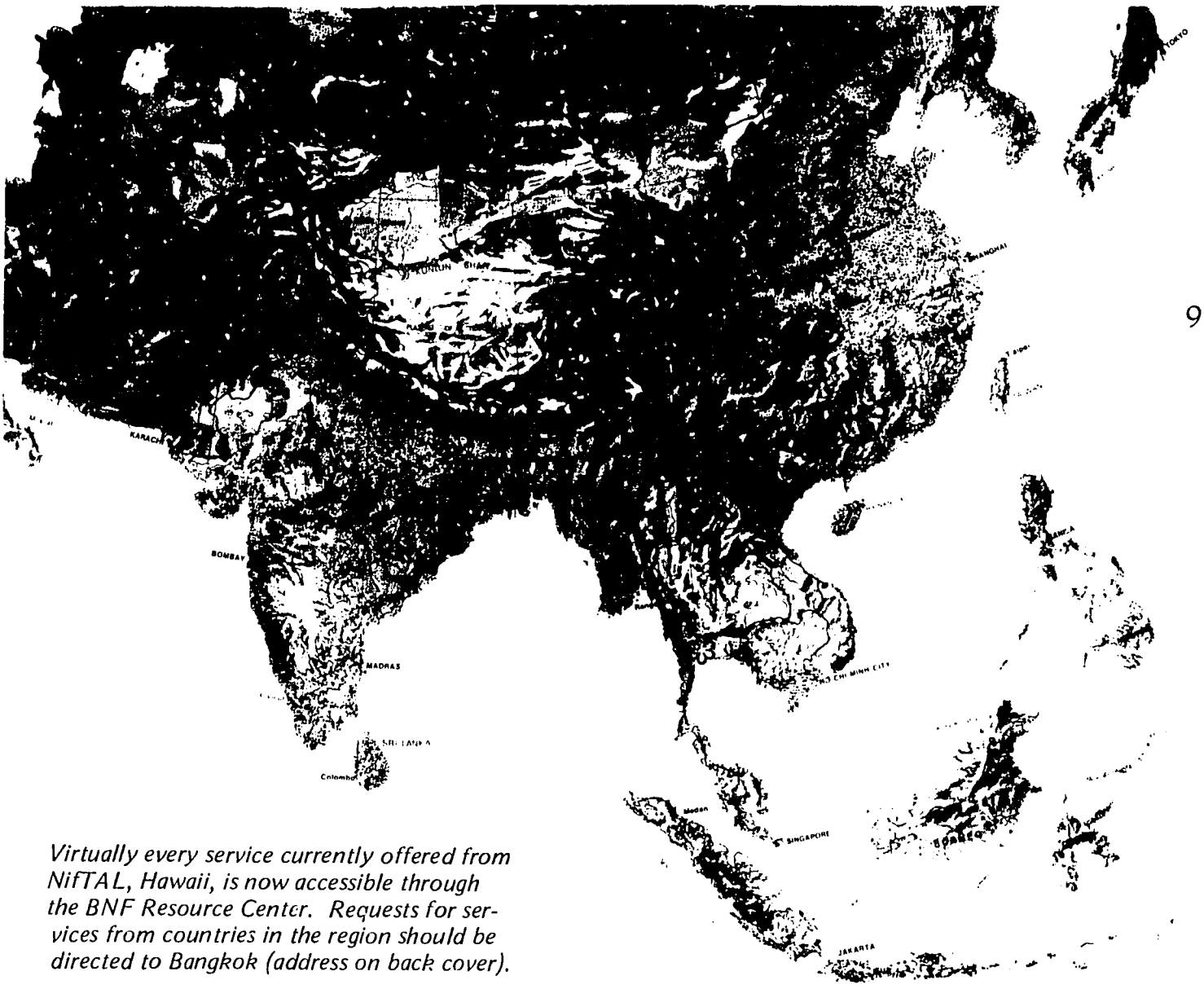
Bangkok was considered the most appropriate location for the Center, because of Thailand's pivotal position in the region and a longstanding tradition of cooperation between the University of Hawaii NifTAL Project and the Soil Science Division of the Thailand Department of Agriculture. The Department 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, a planning workshop was held in Bangkok to devise a workplan for the BNF Resource Center matched to the priorities for biological nitrogen fixation research and production for existing countries of the region.

Scientists and administrators presented synopses of BNF programs in their countries. Representatives detailed the most pressing research and promotional needs to put BNF to work in farmers' fields.



The mainstay of the BNF Resource Center's activities is the provision of technical assistance. The BNFRC, headed by Executive Director Dr. Doug Beck (pictured above), also offers research tools, skills, and knowledge that will enable regional scientists to be self-sufficient.



Virtually every service currently offered from NifTAL, Hawaii, is now accessible through the BNF Resource Center. Requests for services from countries in the region should be directed to Bangkok (address on back cover).

A steering committee has been formed. It will meet yearly to evaluate past activities and orient the Center toward changing regional needs. This committee will be composed of two elected delegates—one each from a country of South and Southeast Asia, the Director General of the Thai Department of Agriculture, Head of the Thai Soil Microbiology Branch, the Director of the Resource Center, and the NifTAL Director. Regional delegates, who will be chosen by their peers, will serve a one year term and will represent the Center at various functions, as well as help to guide the operation of the Center.

The first in-country assignment for the BNF Resource Center is to mount a BNF Workshop for Senior Administrators and Extension Leaders. The workshop, requested and funded by the Bangladesh Agricultural Research Council, is slated for March 1984.

Objective

— to disseminate NifTAL research output, and transfer professional skills and technical information for research, extension, and production enterprises required to assess and use technologies based on biological nitrogen fixation for crop production in the tropics.

Accomplishments

NifTAL research output is disseminated through journal publications and conference/workshop presentations. Output for 1983 is detailed in Appendix I. All papers from research supported by NifTAL since 1976 are listed in Appendix II.

During 1983 there were 378 requests for documents authored by NifTAL staff, and 225 copies of the proceedings of the CIAT/ICRISAT/NifTAL workshop on BNF Technology for Tropical Agriculture were distributed.

Past and current technical information on legume nitrogen fixation is available to researchers in developing countries through the NifTAL Document Collection. This collection now has 7,000 entries, of which just over 1,000 were added in 1983. Two bibliographies (1978 and 1980) and a periodic bibliography (inserted in the BNF Bulletin) detail the documents available. During 1983, NifTAL serviced 748 requests for documents from researchers in 44 countries.

NifTAL's BNF Bulletin sustains awareness by a broad international development audience of projects in progress worldwide that impact on the use of biologically fixed nitrogen in agriculture. The Bulletin is also a vehicle for exchange of ideas between researchers and decision makers. Over 1,600 readers in 100 countries receive three issues of the Bulletin annually.

NifTAL continues to target training at professional and technical research personnel. This year, the need to train extension leaders and inoculant producers was also addressed.

Training opportunities for professionals include graduate assistantships to pursue the Ph.D. and M.S. degrees, and short-term internships at NifTAL headquarters covering advanced techniques for applied research.

Junior researchers and technical personnel can apply for intensive, practical short courses offered by NifTAL in conjunction with host institutes in tropical countries. Currently graduate students working in NifTAL's program are from Bangladesh, Nepal, India (2), Zambia, Liberia, Japan, and the U.S.A.

Scientists from Burma, India, Kenya, Mali, and the Sudan undertook internships with NifTAL in 1983.

NifTAL's training offers the essential combination of agronomic and bacteriological skills to developing country colleagues who will ultimately direct or perform production-oriented research in the tropics.

Short-courses conducted in 1983 were:

Legume Inoculant Training Course.
— mounted in India in cooperation with ICRISAT and FAO/UNEP, and offered to participants from India (9), Sudan (2), Egypt (1), and Indonesia (1).

Symbiotic Nitrogen Fixation in Tropical Agricultural Legumes.
— mounted in Costa Rica in cooperation with University of Costa Rica/CONICYT and offered to participants from Argentina (1), Bolivia (1), Brazil (1), Costa Rica (6), Colombia (1), Dominican Republic (1), Ecuador (1), Mexico (3), Peru (1), and Puerto Rico (1). Other international agencies cooperating were: Unesco, UNEP, ICRO, and IITA.

Assessment of Azolla in Latin America.
— mounted in Peru in cooperation with the National Agricultural Research Institute of Peru, and offered to candidates from Peru (6), Uruguay (2), Brazil (1), Costa Rica (1), Colombia (2).

Azolla Workshop
— mounted in Senegal in cooperation with WARDA/ADRAO, and offered to 16 participants from African countries.

Symbiotic Nitrogen Fixation and Legume Inoculation.
— mounted in Thailand in cooperation with the Thailand Department of Agriculture and North Carolina State University and offered to 45 Thai research and extension managers.

Objective

— to apply the physical and human resources established at NifTAL on USAID central funding to specific projects requested by USAID missions of individual countries, or by other development agencies. Such projects are integrated into NifTAL's program, but are separately funded.

ZAMARE Project

NifTAL participates with University of Illinois Urbana-Champaign, University of Southern Illinois at Carbondale, and University of Maryland, Eastern Shores, in a USAID/Zambia project to support Zambian agricultural research and extension. NifTAL has stationed a soil microbiologist with the Ministry of Agriculture at Mt. Makulu who is responsible for research on soil microbiology in support of the soybean improvement program. NifTAL's staff member in Zambia is backed by technical assistance from NifTAL's Hawaii staff in legume inoculant production and extension guidelines on inoculant use. NifTAL is also training a specialist in biological nitrogen fixation methods through an M.S. program at the University of Hawaii. Accomplishments to date include establishment of a national *Rhizobium* culture collection and testing native strains for nitrogen fixation effectiveness with soybeans. The program is funded by USAID/Zambia.

Memoranda of Understanding are but one means by which NifTAL accommodates a variety of research interests with other organizations.

Along with USDA and the University of Guam, NifTAL is conducting winged bean experiments on Guam and the Northern Marianas Islands.

Indo-U.S. Science and Technology Initiative

NifTAL conducts a collaborative program with the Indian Agricultural Research Institute at Delhi and the University of Agricultural Science at Bangalore. The goals of the project are to establish a National *Rhizobium* culture collection, to improve legume inoculant quality, and select superior rhizobia for pigeon pea *(Cajanus cajan)*. The project is supported jointly by the Indian and U.S. governments with U.S. funding administered by the National Science Foundation and AID.

MIRCEN Project

NifTAL is a designated Microbiological Resources Center (MIRCEN) committed to the preservation of agriculturally important microbial germplasm. A main value of this project to NifTAL is that U.N. funding under the project supports participation by nationals of countries not eligible for USAID assistance in NifTAL programs. This activity is conducted under the auspices of Unesco, UNEP, and ICRO with funds administered by Unesco.

Measuring biomass is just one aspect of NifTAL's model seed production nursery program.

Pacific Island Winged Bean Project

There has been a recent upsurge in interest in the winged bean (*Psophocarpus tetragonolobus*) as a vegetable and fodder crop for small holder farmers in the Pacific Basin. NifTAL is conducting experiments in Hawaii, Guam, and the Northern Marianas to develop appropriate rhizobial inoculants for winged bean varieties introduced to differing soil circumstances in the Pacific Islands. This project is funded by USDA CSRS.

Leucaena Seed Production

Demand for seed of the fast-growing, multipurpose tree "leucaena" has led to distribution of some lines of dubious purity and quality. NifTAL has established model seed production nurseries at several sites in Hawaii (and also in Costa Rica and Sierra Leone) to facilitate dissemination of pure lines of preferred leucaena germplasm. This project is funded by NAS through a Donner Foundation Grant.

Nitrogen Fixing Tree Germplasm

An integrated approach is being followed in this project to derive seed/rhizobia/mycorrhizae sets for nitrogen fixing trees with a potential role in development. Support is from the Science Adviser's Office, USAID.

Other projects

In addition to the funded projects already referred to, NifTAL is involved in many joint projects on a cost-shared basis. Organizations working with NifTAL in this mode in 1983 included: FAO, CIAT, UNDP, IADS, ICRISAT, BOSTID, and the Australian Development Assistance Bureau.

Future project opportunities

NifTAL remains responsive to requests from organizations with an interest in applying NifTAL resources to in-country needs that may be more specific than can be accomodated within the framework of NifTAL's current general contract. Requests for participation by NifTAL staff in discussions on the possibilities for such country projects are welcome.

Objective

— to determine, under realistic field conditions, whether the yields of tropical legumes can be increased by inoculating them with selected strains of *Rhizobium*.

Accomplishments

Results from a total of 110 additional INLIT "A" experiments from 18 countries had been received at NifTAL, analyzed during 1983, and the data analyses returned to the cooperators.

Composite analyses on a worldwide basis have been performed on five crop species: *Cicer arietinum, Glycine max, Lens culinaris, Vigna mungo,* and *Vigna radiata.* The results so far indicate that soybeans *(Glycine max)* have the most consistent evidence of a response to inoculation, being directly associated with prior plantings and inoculation of the crop. Trends are also becoming evident in other species which, with more data from additional locations, will permit identification of factors governing whether or not a field inoculation response is to be expected.

Specific services offered through INLIT are: detailed field trial procedures, improved rhizobial germplasm, research grade inoculants, data analysis, and national research program design assistance.

Asian Network

Data from an additional forty-two trials have been returned to NifTAL during 1983, bringing the total of completed trials close to 200. NifTAL has received over 900 requests to conduct trials and the materials required for these have been dispa'ched.

African Network

Thirty experiments are being undertaken by cooperators in African countries. Data from eight completed trials has been received by NifTAL and analyzed.

Latin American Network

A total of 48 trials have been sent to 14 countries in Latin America and the Carribean. One "B" and 12 "A" trials have been completed and data received.

Mid-East Network

Cooperators in 5 Mid-Eastern countries have been sent 13 "A" experiments. Data from three of these trials has been returned and this data has been analyzed.

Services

NifTAL professionals travel regularly in service of researchers cooperating in the INLIT program. Such site visits are also an opportunity for national researchers to draw on the expertise of NifTAL professionals in aspects of agricultural research not directly relating to the INLIT program itself. Country visits also provide an opportunity for NifTAL staff to support USAID Missions.

Specific services offered through INLIT are:

— detailed field trial procedures

— improved rhizobial germplasm

— research grade inoculants

— data analysis

— national research program design assistance.

More than 200 scientists in the third world participate in the INLIT program. Over 300 trials have been analyzed and many of these confirm that inoculation increases yields.

(Top) NifTAL assumes an active role in INLIT to insure that each cooperator has the necessary support services. (Bottom) Each inoculant developed for INLIT contains three serologically distinct, effective strains of Rhizobium *from diverse geographical and host-germplasm backgrounds.*

Objective

— to develop systems for improved inoculant delivery, and for ensuring dependable, effective inoculation in the field.

Background

NifTAL recognizes that the nature of present legume inoculants and the manner in which they are produced are constraining fuller use of the technology by farmers.

Many tropical countries do not have peat suitable for inoculant carrier, and in any case this form of inoculant is highly vulnerable to damage from high temperature exposure during distribution, storage, and use.

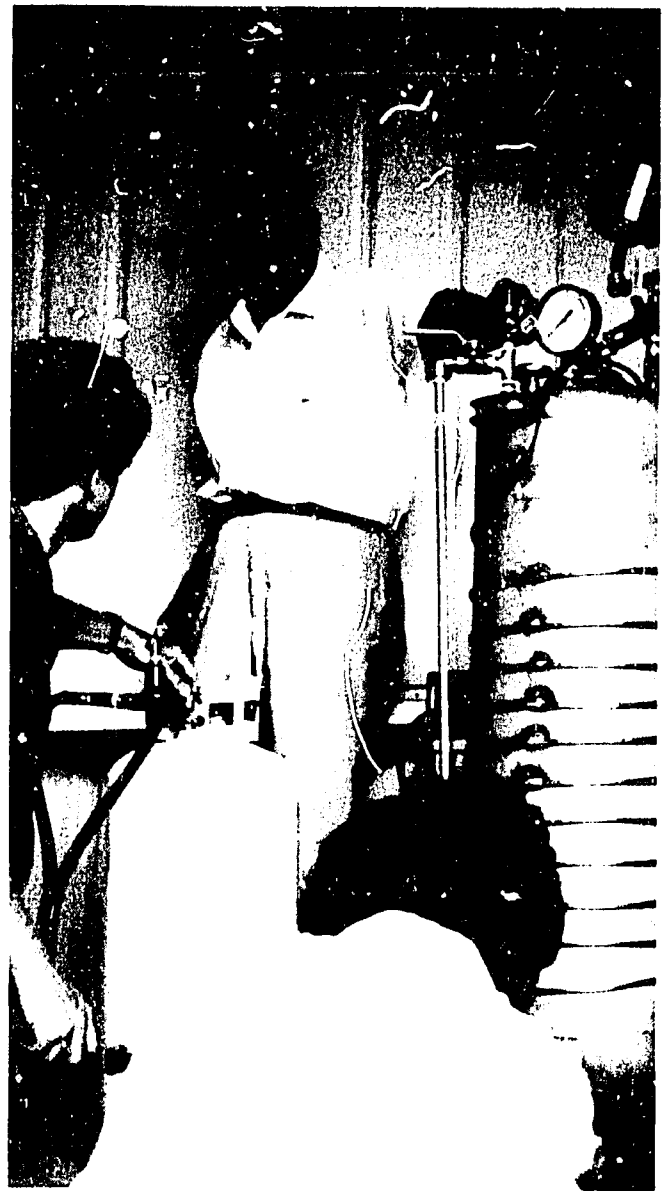
Conventional approaches to inoculant production appear overly sophisticated, risky, and highly capital intensive to entrepreneurs contemplating production in developing countries.

Accomplishments

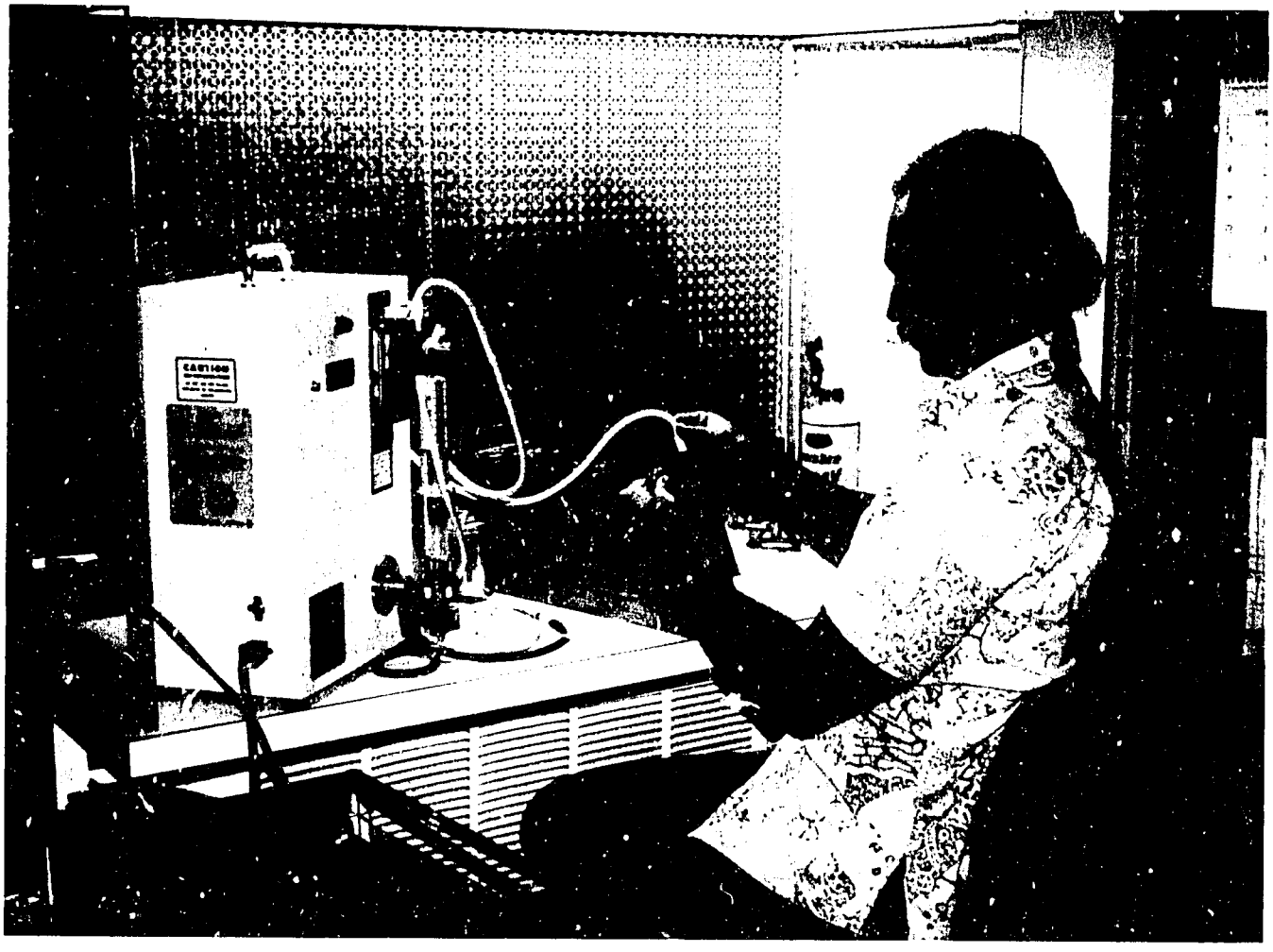
Progress at NifTAL on improving the nature of inoculants has been primarily through development of inoculant strains which exhibit high temperature tolerance and satisfactory shelf storage without refrigeration.

Conceivably, if small-scale, simple, low-cost inoculant production systems were available, inoculants would be produced locally and seasonally, tied to farmer demand. Thus, the major risks to inoculant quality, namely transportation and storage, would be overcome, and the need to change the nature of inoculant would be alleviated. For this reason, NifTAL's main thrust to improve the quality of inoculant reaching farmers is the development of small-scale, low cost production systems.

Previously we reported validation of a dilution approach to eliminating the need for large fermentors. Recent research confirms that the dilution procedure is acceptable using autoclaved peat as a carrier. A pilot version of a shaker based production system has been designed and is operating with excellent results. The system is based as far as possible on



Legume inoculation technology has still to make its full impact felt in agriculture in the developing countries. To meet this challenge, NifTAL is developing low-cost, simple, small-scale inoculant production systems and exploring alternative inoculant carriers.



NifTAL is improving the nature of inoculants primarily through development of inoculant strains which exhibit high temperature tolerance and satisfactory shelf storage without refrigeration.

materials readily available in developing countries rather than specialized items often used in U.S. laboratories, e.g., avoidance of the use of low density polypropylene packages in favor of common polyethylene bags.

Development of low cost fermentors of varying scales has been completed and these are being installed for operation at a remote facility without previous microbiological capability. This experience forces attention to detail, fine tuning the inoculant production technology packages on offer from NifTAL.

After due care is taken to ensure that quality inoculants reach farmers, much still depends on how cautiously the inoculant is handled and how competently it is applied. A pocket manual for extension personnel was therefore prepared giving background on legume inoculants and detailing in plain language how they should be used. FAO joined with NifTAL in this venture and the pocket book will be available early in 1984.

Inoculant Development Services

— technical assistance on inoculant production systems of differing capital costs.

— advisory services on strategies for inoculant manufacture, distribution, and quality control.

Objective

— to maintain a *Rhizobium** germplasm resource comprised of authenticated, characterized rhizobial strains from a wide array of plant hosts and diverse geographic origins, from which to select strains which nodulate agriculturally important legumes under conditions prevailing in the tropics.

Accomplishments

The culture collection was increased only slightly this year, reflecting current policy to add only strains that significantly improve the diversity of the resource. Some 44 new strains for nitrogen fixing trees were added, as were 82 strains for legumes of importance in tropical highlands. Among other strains added were 160 Pacific Island strains for winged bean, an underexploited legume receiving increasing attention.

The collection now has 1,774 strains of *Rhizobium*. The strains were isolated from 285 different legume species and were collected in 56 countries.

Concern over long-term stability of cultures prompted transfer to lyophilized storage in sealed glass ampoules. Previously, cultures were stored using the porcelain bead method. Over 30% of the collection has been lyophilized, priority having been given to tested strains recommended for inoculating agriculturally important legumes.

Strains have been acquired and characterized for the 50 target legumes regarded as having a significant role in development. Additional field testing is planned, particularly for tree legumes.

Research quality inoculants continue to be produced and are in demand around the world because: they are prepared using gamma-irradiated peat and present virtually no quarantine risk during international distribution; they contain sets of three or more antigenically distinct strains of rhizobia which can be followed in competition and persistence studies in soil; and they contain high numbers of well characterized rhizobia.

Rhizobium *strain selection is performed to insure that a legume seed inoculant contains a strain, or strains, of* Rhizobium *capable of forming fully effective, N_2-fixing nodules on legume species for which it is recommended and under the conditions of soil and climate in which the legume crop will be grown.*

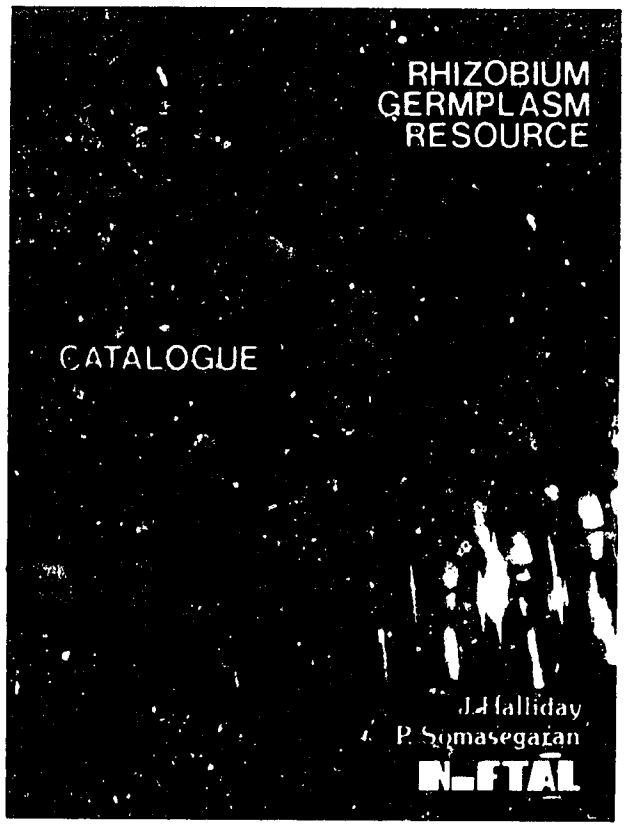
Review of cultures and pertinent information was completed this year. A computerized, data base management system was designed for storage and retrieval of strain data, generating a strain catalogue, and handling the informational services offered to users. Data entry has been completed and a Catalogue of Strains is available.

* *Recently,* Rhizobium *was divided into* Rhizobium *and* Bradyrhizobium. *In this report,* Rhizobium *can be read as* Rhizobium *or* Bradyrhizobium *depending on your acceptance and understanding of the new taxonomy.*

NifTAL serviced over 500 requests for cultures from scientists in 22 countries this year. Culture requests doubled over last year's level.

The MIRCEN Connection

The *Rhizobium* germplasm resource at NifTAL was established with USAID contract funds. This support has been generous but is best viewed as temporary. The University of Hawaii is preserving the collection in perpetuity as a commitment to USAID and with UNEP and Unesco, under the Microbiological Resources Center (MIRCEN) concept. The *Rhizobium* Germplasm Resource at NifTAL is a member of the World Federation for Culture Collections (WFCC) and cooperates with other curators.



A comprehensive Rhizobium *collection has been assembled at NifTAL as a first step in the selection of superior strains to be recommended as inoculants for economically important legume crops.*

The Rhizobium *Germplasm Resource is preserved in perpetuity by NifTAL as a commitment to USAID under the Microbiological Resources Center (MIRCEN) concept.*

Germplasm Services

- catalogue of strains
- *Rhizobium* cultures
- research grade inoculants
- custom carrier supply
- strain data interchange
- custom data searches
- nodule isolations
- long-term repository
- data base design assistance

Enquiries about services are welcome.

Objective

— to establish a serum bank housing antisera for identifying the strains of *Rhizobium* recommended for the economically important legumes under test in the International Network of Legume Inoculation Trials (INLIT).

Accomplishments

A total volume of approximately two litres of antisera has been produced to replenish stocks depleted by requests and to increase the overall volume of reserves held in the collection. The serum bank has been expanded to include six additional antisera for three strains of winged bean rhizobia.

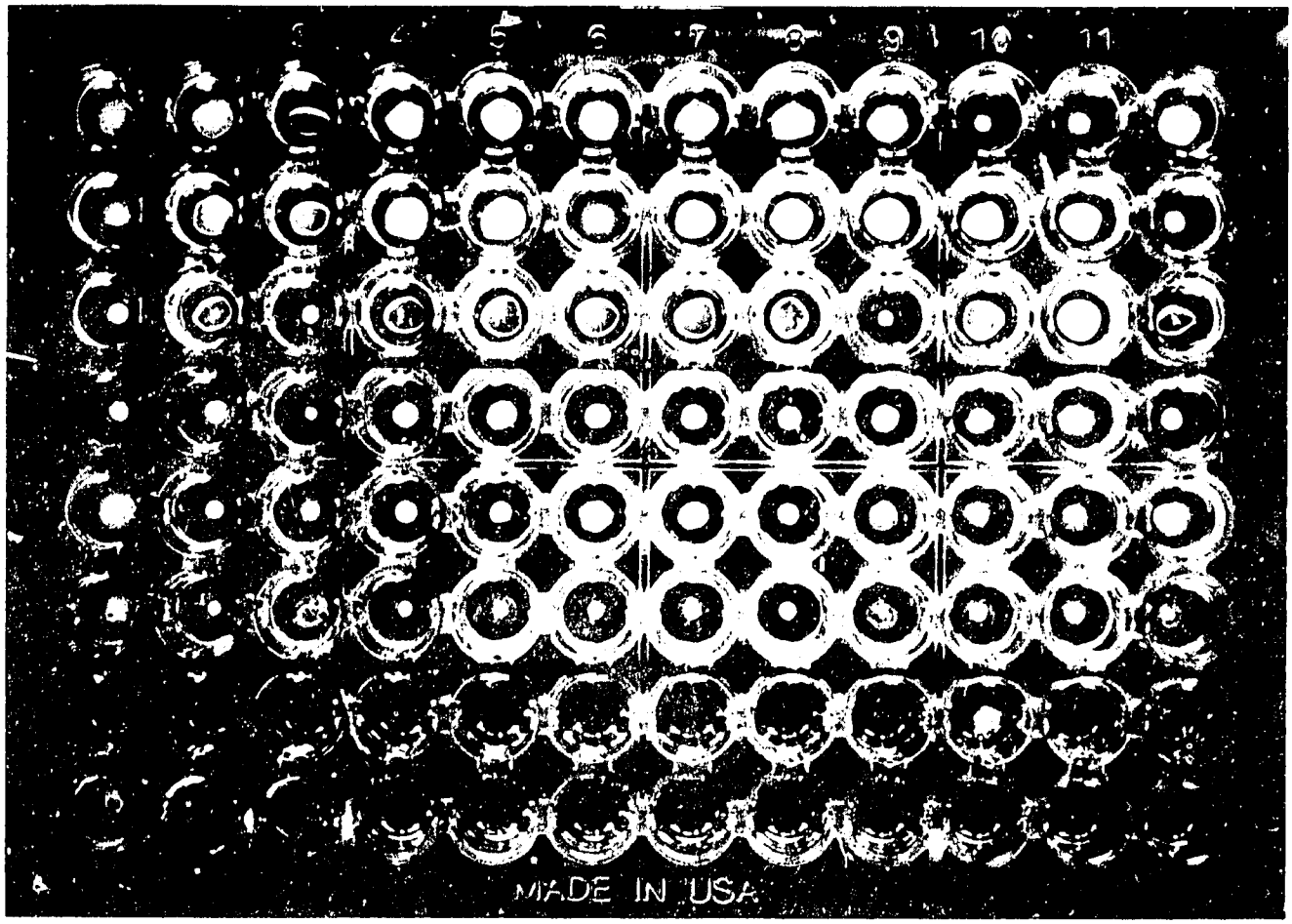
The use of matched Rhizobium *strains and antisera allows a researcher to distinguish introduced rhizobia from all others. This is an essential prerequisite for competition and persistence studies.*

Backed by NifTAL's serum bank, researchers can add a new dimension to field inoculation trials in the tropics.

Approximately 300 ml of goat anti-rabbit globulin (GARG) have been produced for use with the indirect fluorescent antibody (FA) technique. One third of the GARG has been conjugated with fluorescein isothiocyanate (FITC). This GARG-FITC conjugate is now being used for strain identification with all antisera in the NifTAL serum bank.

A computerized data base management system for handling antisera records has been designed. Data input for the list of antisera available through the serum bank has been completed. Cooperators in the INLIT program have been made aware of the antiserum service through the INLIT procedures manual and in the Bulletin to INLIT Cooperators.

Requests for antisera increased markedly this year. A total of 311 ml (in 1 ml aliquots) were supplied to researchers in 13 countries in 1983.



NitTAL offers a variety of antisera services such as custom antisera, antisera for agglutination and gelimmunodiffusion, and an antisera catalogue and manual.

Key role of antisera

One serious obstacle to fuller use of legume inoculation in developing countries is that selected "superior" inoculant rhizobia are often outcompeted by indigenous soil rhizobia. Inoculant strains can fail to establish in the nodule population, and fail also to persist in the soil microflora. Progress cannot be expected until there is a better understanding of the nature of competitive interactions. The use of matched strains and antisera allows a researcher to distinguish introduced rhizobia from all others. This is an essential prerequisite for competition and persistence studies.

It is often not feasible for researchers doing field inoculation trials to produce their own specific antisera. They may lack the facilities, the equipment, or expertise to produce antisera. Yet the use of antisera in research is relatively straightforward, needing only simple materials. Backed by NitTAL's serum bank, researchers can add a new dimension to their interpretation of field inoculation trials performed in the tropics.

Antisera services will continue to expand and a manual of methods for producing and using antisera, and listing antisera on offer from NitTAL, will be available in 1984.

Antisera services

- antisera for agglutination
- antisera for gel immunodiffusion
- fluorescent antibodies
- antisera for fluorescence
- antisera catalogue
- antisera methods manual
- custom antisera
- technical advisory services

Objective

— to derive an improved understanding of how to manage tropical cropping systems to optimize the benefits from nitrogen fixed biologially.

Background

Tropical cropping systems with legumes have traditionally been managed for the non-legume component. Very little is known about if, how, when, and why the cropping system affects nitrogen fixation and/or growth of the legume.

Do legumes even fix nitrogen in mixed cropping systems? How much? If none, why not? What happens to the nitrogen from legume residues? How much is available to subsequent crops? If none, why not?

These are some of the questions that must be answered to maximize the benefits of BNF in tropical cropping systems. Studies carried out under the cropping system objective are designed to answer these and similar questions.

Accomplishments

In July, 1983, NifTAL sponsored a workshop at the Rodale Research Farm in Pennsylvania to review what was known about the nitrogen dynamics of tropical cropping systems and to identify research priorities for NifTAL's cropping system program. Ten experts, representing disciplines as varied as cropping systems, ethnoecolgy, and mathematical modeling, attended.

NifTAL's objective is to develop an improved understanding of how to manage tropical cropping systems to optimize the benefits of nitrogen fixed biologically.

Of great research interest to NifTAL is the mixed cropping system utilizing legumes. It is the dominant cropping pattern in much of the developing world.

They concluded that a dynamic model of nitrogen in tropical cropping systems would be a potentially powerful research and management tool, but the data presently available was insufficient to construct such a model. More information on major nitrogen inputs and outputs was considered necesssary for the development of nitrogen management strategies. Highest priority was given to quantifying BNF and its importance in cropping systems.

Future studies will be mounted in mixed cropping systems. The rationale for this was the dominance of the mixed cropping system in much of the developing world and the scant attention that has been given to this important agricultural system.

Considerable time was devoted to the methodologies available for measuring major nitrogen inputs and outputs.

A questionnare addressing many of the same topics discussed at the Rodale Workshop was distibuted to scientists from 10 Asian countries who attended a later workshop at the Resource Center in Bangkok, Thailand. Some divergence occurred between the Rodale and Bangkok groups on appropriate methodology for mesuring BNF but rankings of research priorities were similar.

Services

This is a new undertaking by NifTAL and services at the present time are limited to the professional expertise of NifTAL staff, primarily in nitrogen cycling in agroforestry systems and legume-based, tropical pastures.

In the near future, the proceedings of the NifTAL-sponsored Rodale Workshop to appraise the state of knowledge on BNF in tropical cropping systems will be available.

Objective

— to lower the cost of soil management, improve crop performance, and guide the choice of legumes for cropping systems by defining their tolerences to components of soil infertility.

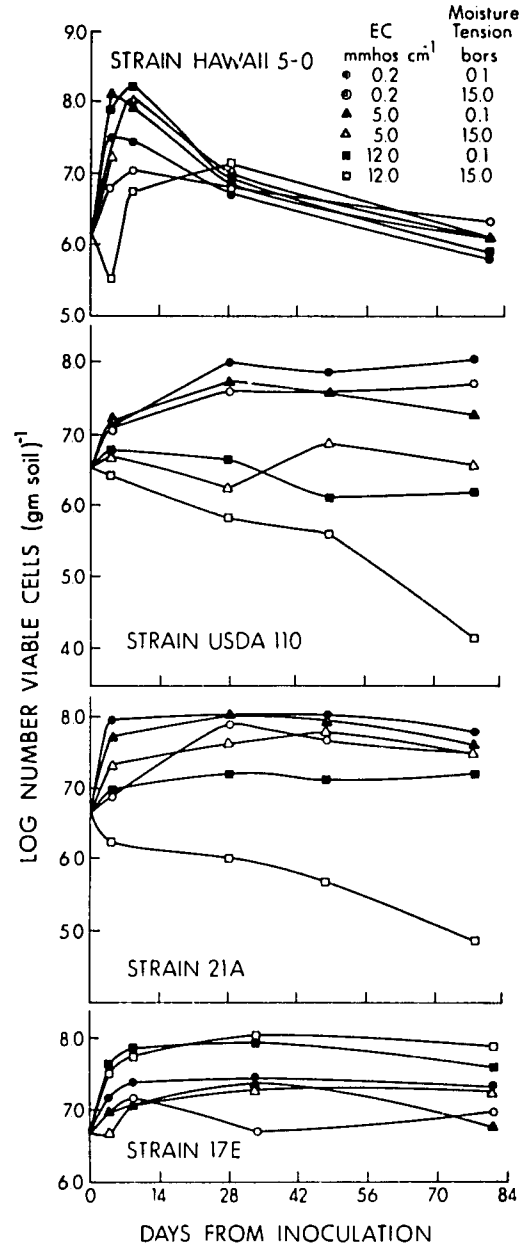
Accomplishments

Research continues on the effects of soil acidity factors on symbiotic performance of legumes. Emphasis was placed on phosphate deficiency and salinity/sodicity stresses that have been neglected in research on symbiotic nitrogen fixation in the tropics. Legumes under study include forage, cover, and woody species likely to be placed into poor soils. Soil fertility requirements of these legume species are essentially unknown. The project extends to selecting rhizobia or plant types with improved tolerance when there is data to indicate that this might lead to worthwhile improvement in the field.

Two cultivars of *Phaseolus lunatus* were found to have high salt tolerance, resembling that of alfalfa or berseem; and six cultivars tested had moderately high acid tolerance, similar to that of cowpea cultivar TVu1190. Preparations were completed for salt screening several species in solution cultures under greenhouse conditions. An outdoor, soil microplot culture system has been devised to screen plants for phosphate tolerance.

A survey of the phosphorus efficiency (= phosphorus required to sustain growth) of 40 strains of rhizobia of various groups was completed. A study with *Rhizobium meliloti* showed that there is interdependence of calcium and phosphorus requirements in these rhizobia. (See Appendix I, Munns.) Most rhizobia performed well at extremely low phosphorus levels. A rapid screening procedure was devised for detecting strains of rhizobia with an abnormally high phosphorus requirement.

Salt sensitivity in *Cicer arietinum* was shown to arise from inability to exclude sodium and calcium from leaves, particularly when plants were grown symbiotically or at low humidity. Chickpea appears to have little control over transpiration or salt translocation due to water loss from glandular hairs. Two less sensitive cultivars were identified, however.



Scientific data like that above are the primary output from the soil stress objective. Further details will be found in NifTAL's many publications listed in Appendices I and II.

NifTAL's objectives in studying soil stress tolerance are to lower the cost of soil management, improve crop performance, and guide the choice of legumes for cropping systems by defining their tolerances to components of soil infertility.

The possibility that extracellular polyssaccharide (EPS) might protect cells from aluminum and acidity by helping to buffer the microenvironment was tested. Representative tolerant and sensitive strains of rhizobia from six groups, and a group of 20 strains of *R. phaseoli* were assayed, but acid tolerance and EPS production were only weakly correlated ($r^2 = 0.33$).

A split-root technique has been used to advantage for distinguishing between host plant-mediated effects of stress treatments and other, more direct effects on *Rhizobium* multiplication, infection, and root nodule function. The studies indicate a greater degree of host plant control over the components of nodulation and nitrogen fixation than have been generally appreciated. Even the outcome of competition between strains of *Rhizobium* for infection sites on legume roots appears subject to pronounced host plant control. These findings underscore the importance of retaining symbiotic competence in improved legume crop varieties.

Soil stress research services

— An array of defined test media and pH indicators for rapid, low cost screening of rhizobia for stress tolerance have been developed. These methods are suitable for laboratories with limited facilities and resources.

— Several experimental systems have been designed for split-root studies. These systems have purposely been improvised from readily available, low cost materials and may bring this valuable technique within the capability of researchers unable to use it previously.

— Selected highly tolerant germplasm identified by the techniques referred to above is available.

Objective

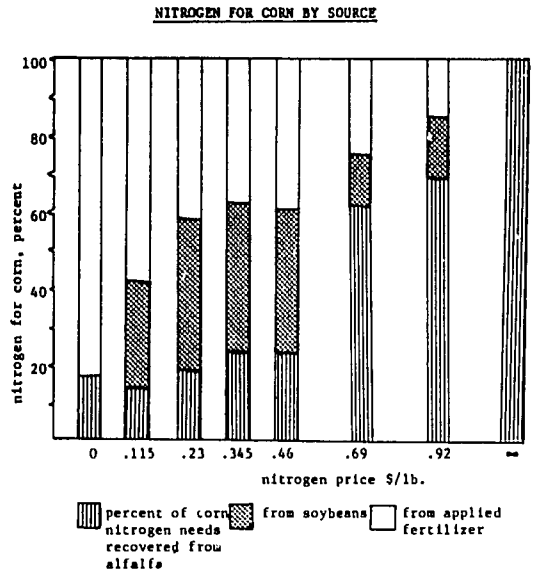
— to devise and test appropriate methodology for measuring the economic benefits of the use of inoculated legume crops in farming systems of developing countries.

Accomplishments

A polyperiod mathematical programming farming systems simulation model was constructed. The model incorporated cropping, livestock, and household subsystems. A large number of alternative crop sequences (rotations), each with several alternative planting and harvesting dates and completely different yields, can be considered simultaneously. Crop output can be sold or eaten, and livestock feed can be raised or purchased.

Biological nitrogen fixation and carryover effects on yield of subsequent crops must be estimated independently, as must fertilizer response functions.

The small bag of inoculant will inoculate soybeans at a fraction of the cost of the large bags of nitrogen fertilizer; and, it will provide the same amount of nitrogen to the crop.



NifTAL has a subcontract with the University of Minnesota to devise an appropriate methodology for measuring the economic benefits of the use of inoculated legume crops in farming systems of developing countries.

The model permits parameterization of key variables, such as estimating the effect of a wide range of nitrogen fertilizer prices or use of legumes in a farming system. The model was tested with data from a southeastern Minnesota alfalfa-based dairy farm system. At any nitrogen fertilizer price above zero, the forage legume was very stable in the farming system studied. Grain legumes were stable at current prices. The model appears now to be ready for testing and use in developing countries.

Future plans

The next phase of this activity involves an in-country study to test and appropriately modify the model to cope with parameters encountered in developing countries. Thailand, and specifically its Northeast region, has been selected for the field study. This is primarily because of strong, long-standing linkages between NifTAL/University of Minnesota and agricultural economists/BNF specialists in Thailand. Thailand is also one of few tropical countries where adequacy of baseline data approaches the level required to conduct the study.

A research assistant is being posted at Khon Kaen University attached to the Farming Systems Project within the Faculty of Agriculture to accomplish on site research.

APPENDIX

Published Papers

- Ayanaba, A., Asanuma, S., and D. N. Munns, 1983. An Agar Plate Method for Rapid Screening of *Rhizobium* for Tolerance to Acid-Aluminum Stress. Soil Sci. Soc. Am. J. 47:256-258.

An agar plate method has been developed for the rapid, preliminary screening of large numbers of rhizobia for tolerance to acidity and aluminum (acid-Al). Media containing pH indicators are used, and distinguish true tolerance from cases where growth depends on a previous pH shift. Strains which were highly tolerant or sensitive to acid-Al on agar behaved similarly in defined liquid media. Strains that formed dry, pinpoint colonies were more sensitive to acid-Al than those that formed large, gummy colonies.

- Beck, D. P., and D. N. Munns, 1984. Effect of Calcium on the Phosphate Nutrition of *Rhizobium meliloti*. Soil Sci. Soc. Am. J. In Press.

- Beck, D. P., and D. N. Munns, 1984. Nodulation of Legumes in a P-deficient Soil as Affected by P-efficiency of *Rhizobium*. Soil Sci. Soc. Am. J. In Press.

- Beck, D. P., D. N. Munns, and K. G. Cassman, 1983. Phosphate Nutrition of *Rhizobium* sp. Appl. Environ. Microbiol. 47 (2): 278-282.

This study was conducted to determine the behavior of 40 strains from six species of *Rhizobium* in liquid defined media containing orthophosphate at levels likely to be encountered naturally, ranging from the high concentrations expected in nodules and artificial media to the low concentrations of soil solutions. Storage capacity in strains with high levels (2mM) of P and ability to utilize this stored P for growth after transfer to low levels (0.06 uM) of P varied with each strain. Storage varied from about 1 to 2% P (dry weight) for all strains, with the number of generations supported dependent on the quantity of P stored and on the utilization efficiency. The ability to store P at high levels is probably less important than the uptake and utilization efficiency of P supplied at low levels. Strains varied greatly in tolerance to low levels of P maintained in solution by an iron oxide buffering system. Differences in growth rate at low levels of P were large enough to be agronomically important.

- Bohlool, B. B., Kosslak, R., and R. Woolfenden, 1984. The Ecology of *Rhizobium* in the Rhizosphere: Survival, Growth and Competition. In C. Veeger and W. E. Newton (Eds.) Advances in Nitrogen Fixation Research. Martinus Nijhoff/Dr. W. Junk Publ., The Hague, p. 287-293.

Quantitative ecology of desired strains of *Rhizobium* in soil, rhizosphere and nodules can be studied directly using strain-specific fluorescent antibodies. Competition among strains for nodulation of the appropriate host is a complex process involving strain/strain, as well as, strain/host interactions. This paper discusses the rhizosphere competence of different strains of *R. japonicum* and *R. leguminosarum* in the rhizosphere of soybeans, peas and corn in relation to nodule occupancies of each strain on the host. Evidence is presented on how the host plant may orchestrate the selection of one strain over another in the early stages of the infection process.

- Burton, J. C., 1983. Legume Inoculants and their Use. FAO, Rome, 85 p. No abstract.

- Burton, J. C., 1983. TDY Report—Legume Inoculant (Rhizobia) Production for Soybeans and Other Legumes in Egypt. EMCIP Publication No. 5, Consortium for Int'l. Development, Publishers, 30 p. No abstract.

- Burton, J. C., 1984. Legume Inoculant Production. NifTAL, Hawaii, 50 p. No abstract.

- DaSilva, E. J., Freire, J. R. J., Keya, S. O., and J. Halliday, 1983. The Role of MIRCEN's in Rural Development, Agricultural Training and Environmental Management. In Training for Agricultural and Rural Development 1982. FAO, p. 23-28. No abstract.

- Davis, R. J., Halliday, J. and F. B. Cady, 1984. Preliminary Data from Worldwide Legume Inoculation Trials. In C. Veeger and W. E. Newton (Eds.) Advances in Nitrogen Fixation Research. Martinus Nijhoff/Dr. W. Junk Publ., The Hague, p. 333.

INLIT is a program initiated in 1980 which constitutes a network of cooperators which are either individual research scientists or institutions. These cooperators are located in developing countries and conduct legume inoculation trials. Cooperators are supplied a standard experimental design, supplies such as an inoculation package (seed if necessary), consultation, training and data analysis. Results from 110 trials has been reported and indicate that Glycine max gives the most consistent evidence of a response to inoculation, being directly associated with prior plantings and inoculation of the crop. However, trends are evident in other species indicating that when more locations have been reported factors will be identified which will indicate whether or not a response can be expected with these species.

- Eriksen, F. I., and A. S. Whitney, 1984. Effects of Solar Radiation Regimes on Growth and N2 fixation of Soybean, Cowpea, and Bushbean. Agron. J. 76, accepted.

Legumes are commonly grown in the tropics under reduced solar radiation due to dense cloud cover and shading from plantation or tall intercrops. This paper examines the growth and N^2 fixation of several grain legumes common to the tropics on an Oxic Haplustoll in Hawaii. Plantings were made in April (warm season) and November (cool season) of soybean (*Glycine max* L. Merr.) 'Kahala'; cowpea (*Vigna unguiculata* Walp.) 'TVu 1190' (April) and 'TVu 4557' (Nov.); and bushbean (*Phaseolus vulgaris* L.) 'Burpee Tenderpod'. Polypropylene netting was used to obtain 27, 45, 70, and 100% of the incoming solar radiation through the entire crop cycle. Consistent shading effects were with the three legumes during both warm and cool seasons even though absolute yields were lower in the cool season due to day length and temperature sensitivity

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in soybean and cowpea. Nitrogen fixation parameters generally followed dry matter (DM) and seed yields. Total nodule activity (TNA) was very low in all the legumes at 27% sun. With the exception of bushbean at 27% sun, shading caused a 34% average reduction in DM and grain yields, vs. a 50% reduction in TNA, and a 76% reduction in nodule mass. Soybean TNA peaked 3 to 4 weeks after flowering, cowpea (cool season) TNA peaked at flowering, and bushbean TNA peaked slightly before or at flowering. There was a tendency for a delay (approx. 2 weeks) at 70, 45, and 27% sun in the final stages of TNA, for summer and cool-season soybean and bushbean. These delays were not noted for cowpea or cool-season bushbean. Of the three legumes, cowpea was the least shade tolerant. It produced more DM at full sun than either soybean or bushbean, but at 27% sun, DM yields were similar for all three crops. The response of soybean to shade was intermediate with no reduction in DM and grain yield at 70% sun. Bushbean was the least affected by shading; in fact, maximum yields were obtained at 70% sun. At full sun, their leaves were yellow and seed weight was low; this problem could be corrected by N fertilization.

- Halliday, J., 1983. Biological Nitrogen Fixation: What's in it for Hawaii? In Proceedings 1st Fertilizer and Ornamentals Workshop. Univ. of Hi. Res. Ext. Ser. 037, p. 75-86. No abstract.

- Halliday, J., 1983. *Rhizobium* Relations of Tropical Legumes in Tropical Soils and Implications for Agrotechnology Transfer. Benchmark Soils Proj. Tech. Note 7, p. 129-140.

A technology is available for establishing effective nitrogen-fixing symbioses between legumes and rhizobia. It involves application of *Rhizobium* bacteria either to the legume seed-coat prior to planting or directly into the soil in which the legumes will be sown. Since specific rhizobial inoculants are marketed on the basis of the legume species and variety with which they are to be used—not based on the characteristics of the soil into which they are introduced—it is inferred that the same inoculant is expected to perform equally well at locations with different soil characteristics. *Leucaena leucocephala* is the only legume for which the inoculant strain varies depending on the characteristics of the soil into which it will be introduced. *Rhizobium* strain CB 81 is recommended for *Leucaena* grown in acid soil and strain NGR 8 for alkaline soil. Some account is taken of the soil type in the inoculating procedure. In general, lime-pelleting of inoculated legume seeds is advocated for plantings in acid soils, though some authorities consider it unnecessary and even harmful to lime-pellet alkali-producing strains of tropical rhizobia. Results from field trials of strains of *Rhizobium* for tropical forage legumes at two sites in Colombia and of rhizobia for *Phaseolus vulgaris* at two sites in Hawaii are used to illustrate the influence of soil and environment on legume response to inoculation and the relative performance of strains introduced to similar and different soils. The feasibility of taking account of soil characteristics in development of inoculation recommendations and the possibility that performance of inoculants could be predicted on the basis of soil taxonomy are discussed.

- Halliday, J., 1984. Integrated Approach to Nitrogen Fixing Tree Germplasm Development. Rev. Bras. Sci. Agropec. (special volume), 21 p.

The performance of nitrogen fixing trees introduced to new environments depends on proper reconstitution of the symbiotic associations on which the trees rely for their nutrition. Thus selection strategies employed to identify adapted germplasm for particular sites must provide for three-way selection of seed, rhizobia, and mycorrhizae. Selected lines must then be multiplied before they can be deployed in varying types of development programs. Special problems are faced in accomplishing these ends with virtually all nitrogen fixing trees. Results and experiences are described which emphasize the importance of parallel selection of plant germplasm and *Rhizobium* strains. In the case of VA mycorrhizae, effective symbioses can occur without specific inoculation. Methods for selecting and multiplying trees and their microsymbionts on a large scale are described and discussed.

- Halliday, J., 1984. Guidelines for Collecting the Root Nodules of Leguminous Trees. In P. Huxley, et al (Eds.) A Manual of Methodology for the Exploration and Assessment of Multipurpose Trees (MPTs). ICRAF, Nairobi. No abstract.

- Halliday, J., 1984. Methods for Assessing Rhizobia and VA Mycorrhizae for Multipurpose Trees. In P. Huxley, et al (Eds.) A Manual of Methodology for the Exploration and Assessment of Multipurpose Trees (MPTs). ICRAF, Nairobi. No abstract.

- Halliday, J., 1984. Principles of *Rhizobium* Strain Selection. In M. Alexander (Ed.) Biological Nitrogen Fixation. Plenum Publ., Corp. p. 155-171. No abstract.

- Halliday, J., 1984. Guidelines for Collecting the Root Nodules of Leguminous Trees. Nitrogen Fixing Tree Res. Reports Vol. 2. No abstract.

- Halliday, J., 1984. Register of Nodulation Reports for Leguminous Trees and Other Arboreal Genera with Nitrogen Fixing Members. Nitrogen Fixing Tree Res. Reports Vol. 2. No abstract.

- Halliday, J., and P. Nakao, 1984. Technical Note on the Germination of Leguminous Tree Seeds. Rev. Bras. Sci. Agropec (special volume).

Seeds of 33 leguminous tree species were tested for germination following mechanical or chemical scarification. The treatments gave germination of over 50% for 28 of the species. Mechanical or chemical scarification of about three times the number of seeds needed for an experiment with leguminous tree species is recommended as a rule of thumb when the germination rate of the seeds is in doubt.

- Halliday, J., and P. Somasegaran, 1983. Nodulation, Nitrogen Fixation, and *Rhizobium* Strain Affinities in the Genus *Leucaena*. Leucaena Research in the Asian-Pacific Region. IDRC, p. 27-32.

Leucaena spp. belong in subfamily Mimosoideae of the Leguminosae. All 10 species have been reported to nodulate readily, but some doubt remains in the case of *L. retusa*, which has failed to nodulate in Hawaii soils. The infection process has not been studied in leucaena. Nodule shape is variable, being astragaloid, desmodioid, or muconoid in form. Attachment to the root is narrow and fragile. Annual nitrogen accumulation rates of 600 kg/ha and as high as 1 t/ha are on record, but annual nitrogen fixation has not been determined with precision. One approximation, based on the acetylene-reduction technique, puts annual fixation at 110 ± 30 kg. The specific designation for the leucaena rhizobia is *Rhizobium loti*. These rhizobia are typically fast growing, gummy, and translucent on yeast-mannitol agar (YMA). They are neutral, or slightly acidic, in their pH reaction on YMA. Leucaena rhizobia will nodulate other tropical legumes usually nodulated by fast-growing rhizobia and will also nodulate some species normally nodulated by slow growers. Slow-growing strains of *Rhizobium* from other tropical genera do not nodulate leucaena. Leucaena usually nodulates spontaneously with indigenous soil rhizobia without specific inoculation, but the documented cases of nodulation failure are sufficient to prompt adoption of legume inoculation when leucaena is introduced to locations where it has not been grown before. Although leucaena performs very poorly in acid-soil sites, strains of *Rhizobium* for leucaena are available that exhibit tolerance to low pH. Acid tolerance in a strain is not dependent on an alkali-producing capability. One particular strain, TAL 1145, has proven to be competitive for nodule formation, persistent in the soil microflora, and fully effective as a nitrogen-fixing partner for leucaena in acid soils.

- Halliday, J., and P. Somasegaran, 1984. The *Rhizobium* Germplasm Resource at NifTAL Catalogue of Strains. NifTAL Project, 94 p. No abstract.

- Kilkenny, M. R., 1984. An Economic Assessment of Biological Nitrogen Fixation in a Farming System of Southeast Minnesota. M.S. Thesis, Univ. of Minn. 215 p.

- King, J. W., 1983. Pretesting Communication Materials in International Agricultural Settings. Agric. Information Dev. Bull. 5 (4): 13-17.

Although the quality of any communication effort is based on the quality of decisions regarding that effort, these decisions are built on identifying alternatives and on making judgments about those alternatives. Judgments are based on available information. The means necessary to provide this information for decision making comprise the area of evaluation. One particular type of evaluation is pretesting or formative evaluation. Pretesting has been discussed in international agricultural communication, it has not been implemented to the extent that it could be. Pretesting is an evaluation process that includes several activities aimed at collecting and interpreting information for possible revision and improvement of communication efforts. Pretesting can be done in two phases, first to review communications plans before they are implemented and secondly to revise communication products before they are developed and disseminated. Concepts, objectives, scripts, partially completed materials, alternative versions, and final products can all be pretested to determine their potential effectiveness. The variables of effectiveness covered by pretesting include attraction (attention, interest, appeal, enjoyment), comprehension (readability, understandability), acceptability (credibility, relevance, believability, identification), self involvement (personal relevance), recall (memorability), and persuasion (do-ability). Several simple techniques can be used to pretest materials. These include draft reviews, storyboard testing, stop methods, one channel methods, observations, group discussions or interviews, and questionnaires.

- King, J. W., 1983. Revising Training Media. Performance and Instruction J. 22 (4): 9-10.

This paper presents a model for revising training materials. As training and instructional materials become dated, there is usually a need to produce new materials. These updated products will both revise and enhance old content. Revision of materials can be divided into six steps: 1) previewing the materials; 2) developing a revision strategy; 3) developing the questions; 4) presenting the materials to be revised to a test audience; 5) collection and summarizing the data, and making recommendations; and 6) developing recommendations. A thirteen item assessment list with corresponding questions is them presented.

- King, J. W., 1984. Large University Agricultural Projects —Communication Problems and Prospects. ACE Quarterly 67 (1): 19-24.

This paper discusses the problems of and prospects for international communication sections of large university agricultural projects. Roles of communication sections included media production, information dissemination, publication, library maintenance, and training. Most sections see themselves as linking mechanisms, translating and tailoring project content for identified special audiences. Problems were identified as "systems" problems (lack of funding, staff turnover, ill-defined audiences, lack of credibility, lapses in intraproject communication, limited interpretation of communication, and inability to reconceptualize roles) and "diffusion" problems (inadequate unsystematic structure, lack of openness, inadequate capacity, lack of reward, distance and lack of synergy). Prospects for communication sections reflected communication opportunities (systems approaches, network funding and emerging characteristics of future-oriented sections) and individual approaches (professionalization of communication, need to be proactive and continuing education).

- May, S. N., and B. B. Bohlool, 1983. Competition Among *Rhizobium leguminosarum* Strains for Nodulation of Lentils (Lens esculenta). Appl. and Environ. Microbiol. 45 (3): 960-965.

Thirty-one cultures of *Rhizobium leguminosarum* were screened for effectiveness (C_2H_2 reduction) of lentils (*Lens esculenta*). Fluorescent antibodies prepared against three of the most effective strains (Hawaii 5-0, Nitragin 92A3 and Nitragin 128A12) exhibited a high degree of strain specificity; the antibodies reacted strongly with their

homologous rhizobia in culture and with bacteroids in nodules. They did not cross-react with one another, and only weakly with 5 of the 47 other *R. leguminosarum* cultures tested. In competition studies in the growth chamber, whenever strain Nitragin 92A3 was included in the inoculum mixture, it consistently (but not always significantly, P = 0.05) occupied the majority of nodules on all four cultivars used. However, some degree of strain X cultivar interaction was apparent: Hawaii 5-0 was of equal competitiveness (P = 0.05) with Nitragin 92A3 on three varieties (Commercial, Tekoa and Benewah), but inferior (P = 0.01), on the Chilean variety; Nitragin 92A3 completely dominated (P = 0.01) Nitragin 128A12 on all cultivars; and Hawaii 5-0 was of equal competitiveness (P = 0.05) to Nitragin 128A12 on the Chilean variety and more competitive (P = 0.01) on the commercial variety and less on the other two varieties. In field experiments, Hawaii 5-0 proved of equal competitiveness (P = 0.01) with Nitragin 92A3 in one soil (an Inceptisol) and superior ($P \leq 0.05$) to it in another (an Oxisol). Incidence of double-strain occupancy of nodules varied from 0 to 36% in vermiculite, depending on the strains in the mixture and the host variety, and from 0 to 38% in the field, depending on the strains in the mixture and the soil type. The results suggest a close relationship between the competitiveness of a strain and its occurrence in doubly infected nodules.

- McNeil, D. L., Borton, S., Amara, D., and M. S. Vora, 1983. Use of Antibiotic-Resistant *Rhizobium* Mutants for Competition Studies with *Cajanus cajan*. Int'l. Pigeonpea Newsltr. 2:71-72.

Antibiotic-resistant mutants of *Rhizobium* for pigeonpea were compared with their parent strains in determining effectiveness and competitiveness in both Leonard jars with sterile medium and pots containing nonsterile field soil. Results indicated that the use of spectinomycin or spectinomycin/streptomycin mutants proved as sensitive as the serological method (FA) for determining nodule occupancy and can give results very similar to those found with serological techniques if background antibiotic resistance of the *Rhizobium* is low.

- Munns, D. N., 1984. Coping with Soil Acidity in Legume Symbiosis. Australian Inst. of Agric. Sci. Occasional Pub. No. 12. No abstract.

- Roskoski, J. P., and T. Wood, 1984. Nodulation and Nitrogen Fixation by Five Species of Leguminous Trees Grown in Soil From Undisturbed and Disturbed Tropical Sites in Mexico. In C. Veeger and W. E. Newton (Eds.) Advances in Nitrogen Fixation Research. Martinus Nijhoff/Dr. W. Junk Publ., The Hague, p. 357.

Leguminous trees that grow rapidly, fix nitrogen, and produce fuelwood and protein-rich foliage and fruits hold potential for use in tropical areas. Many of the valuable characteristics of these plants depend, in part, on the formation of effective nitrogen-fixing symbiosis. Examined was the ability of tropical soils from different sites in Mexico to supply rhizobia to *Acacia pennatula* Schl. and Cham., *Albizia lebbek* L., *Enterolobium cyclocarpum* Jarq., *Gliricidia sepium* Jarq., and *Leucaeana leucocephala* Benth. These species were planted in soil from a primary rain-forest, two slash-burn fields, two pastures, a secondary forest and two eroded sites. Plant height and biomass, and nodular biomass and specific activity were determined on 2 and 6 month old seedlings. The latter were also used to establish $^{15}N_2/C_2H_2$ conversion ratios. Overall, we found that even soils from adjacent areas varied markedly in their ability to support tree legume growth and nodulation, and that conversion of tropical forests to other land uses increased the diversity and abundance of rhizobia specific of leguminous trees.

- Singleton, P. W., 1983. A Split-Root Growth System for Evaluating the Effect of Salinity on Components of the Soybean *Rhizobium japonicum* Symbiosis. Crop Sci. 23:259-262.

Study of the effects of salinity on symbiotic nitrogen fixation is complicated by the high degree of integration between shoot and nodules. Shoot photosynthetic capacity determines not only the amount of photosynthate available to the nodules, but the strength of the sink for nodule products. Salinity may also directly affect functional processes within the nodule. Two experiments were undertaken which employed a split-root solution culture assembly that confined N_2 fixation to one-half of soybean [*Glycine max* (L.) Merr.] root systems. In one experiment, one-half of the root system was inoculated with a non-N_2 fixing strain (ineffective) of *Rhizobium japonicum*, SM-5, 48 hours prior to inoculating the other side with effective strain USDA 110. Nodulation and N_2 fixation by USDA 110 when in combination with SM-5 was significantly suppressed compared to a second treatment where one-half of the root system was inoculated with USDA 110 and the other half remained uninoculated. Root development was significantly greater on the side producing effective nodules than that of the uninoculated or SM-5 nodulated half-root systems. In another experiment, one-half of the root system was nodulated with USDA 110 and the opposite side with SM-5. After nodule development, NaCl (120 mM) was added to the nutrient solutions of: 1) neither half, 2) effective nodule (USDA 110) half, 3) ineffective nodule (SM-5) half, or 4) both halves of the root systems. Osmotic potentials; ($\psi\pi$) of roots, nodules, and leaves were measured. Leaves of plants with only half their roots salinized had $\psi\pi$ (-10.3 bar and -11.1 bar treatments 2 and 3 respectively) comparable to the non-salinized control (-9.3 bar). Leaf $\psi\pi$ of plants with both root halves in salt solutions was -17.9 bar. Nodules exposed to salt solution had values ranging from -9.3 to -12.2 bar while $\psi\pi$ of nodules in normal solutions ranged from -6.1 to -7.8 bar. Ineffective nodules always had a lower $\psi\pi$ than similarly treated effective nodules. These results demonstrate that functional components of the soybean *Rhizobium japonicum* symbiosis can be independently subjected to salinity stress. This methodology can be used to evaluate the relative sensitivity of the components of the symbiosis.

- Singleton, P. W., and B. B. Bohlool, 1983. Effect of Salinity on the Functional Components of the Soybean-*Rhizobium japonicum* Symbiosis. Crop Sci. 23:815-818.

A split-root solution culture system was used to partition the effects of salinity on the functional components of the soybean [*Glycine max* (L.) Merr.]—*Rhizobium japonicum* symbiosis. Nodules were confined to half of the root system. After nodules were well-developed (40 days from planting), 120 mM NaCl was applied to the half-root systems in the following combinations: 1) unstressed, no salt to either half-root system; 2) partially stressed shoot and unstressed nodules, salt to only the nonnodulated half-root system; 3) partially stressed shoot and stressed nodules, salt to only the nodulated half-root system; and 4) stressed shoot and nodules, salt to both half-root systems. Leaf osmotic potentials (ψs) for the four treatments were: 1) -1.02; 2) -1.12; 3) -1.23; and 4) -1.83 MPa. Nodule ψs for the treatments were: 1) -0.70; 2) -0.72; 3) -1.12; and 4) -1.21 MPa. While total and specific nitrogenase activity as well as shoot N content were suppressed in treatment 3, the greatest decrease in activity and shoot N was observed when the plant was stressed by having both half-root systems in salt (treatment 4). The rate of leaf expansion in treatment 4 was half the rate observed in the other treatments. We conclude that reduced N_2 fixation by nodulated soybeans growing in saline environments was more a result of the effect of salt on leaf ψs and expansion, than the direct action of salt on the functional processes of the nitrogenase system.

- Singleton, P. W. and B. B. Bohlool, 1984. The Effect of Salinity on Nodule Formation by Soybean. Plant Physiol. 74:72-76.

A split-root growth system was employed to evaluate the effect of NaCl on nodule formation by soybean. By applying the salinity stress and rhizobial inoculum to only one-half the root system the effects of salinity on shoot growth were eliminated in the nodulation process. *Rhizobium* colonization of root surfaces was not affected by the salt treatments (0.0, 26.6, 53.2 and 79.9 mM NaCl). While shoot dry weight remained unaffected by the treatments, total shoot N declined from 1.26 g N/pot at 0.0 mM NaCl to 0.44 g N/pot at 79.9 mM NaCl. The concentration of N in the shoot declined from 3.75% N (control) to 1.26% N at 79.9 mM NaCl. Nodule number and weight were reduced approximately 50% at 26.6 mM NaCl, and by more than 90% at 53.2 and 79.9 mM NaCl. Total nitrogenase activity (C_2H_2 reduction) declined in relation to nodule number and dry weight. Specific nitrogenase activity, however, was less affected by salinity and was not depressed significantly until 79.9 mM NaCl. Isolates of *R. japonicum* reisolated from nodules formed at 79.9 mM NaCl did not show increased nodulation of roots under salt stress compared to nodule isolates from normal media. The early steps in nodule initiation are, therefore, extremely sensitive to even low concentrations of NaCl. The sensitivity is not related to rhizobial survival and is probably due to the salt sensitivity of root infection sites.

- Singleton, P., and B. B. Bohlool, 1984. The Effect of Salinity on Survival of *Rhizobium*, Nodule Function and Nodule Formation in the Soybean-*Rhizobium japonicum* Symbiosis. In C. Veeger and W. E. Newton (Eds.) Advances in Nitrogen Fixation Research. Martinus Nijhoff/Dr. W. Junk Publ., The Hague, p. 545.

Symbiotic nitrogen fixation may be adversely affected by saline environments. Described are three experiments that assess the salt sensitivity of: 1) *Rhizobium* as free living organisms; 2) soybean nodule function; and 3) soybean nodule formation. In addition, a split-root plant growth system is described which can be used to separate the effects of salinity stress on host yield potential from the effects of salinity on nodule processes.

- Singleton, P. W., and K. R. Stockinger, 1983. Compensation Against Ineffective Nodulation in Soybean. Crop Sci. 23:69-72.

Two cultivars of soybean [*Glycine max* (L.) Merr.] were inoculated with nine ratios of ineffective (SM-5) and effective (CC709) strains of *Rhizobium japonicum*. Plants were grown in the greenhouse and harvested after 33 days. Nitrogenase activity, total shoot N, nodule number, nodule mass, and the proportions of nodules formed by each strain were determined. Nodules were identified serologically by the fluorescent antibody technique. Significant increases in total N in the shoot occurred as both the proportion and number of nodules formed by effective strain CC709 increased. Shoot N was highly correlated with nitrogenase activity and nodule mass. The relationship between percent effective nodules and shoot N was nonlinear so that 95% of maximum N accumulation was obtained when only 75% of the nodules (on a nodule number basis) were effective. The average size of an effective nodule was at least 2.5 times larger than an ineffective nodule and their average weight increased from 2.38 to 4.88 mg/nodule as the proportion of effective nodules declined from 100 to 18.8%. The proportion of total nodule tissue that was effective was greater than the corresponding percent effective nodules. This compensatory mechanism tended to keep the amount of effective nodule tissue constant up to a point even though the proportion of effective nodules declined. There was a deleterious effect on N_2 fixation only when infection by the ineffective strain was great enough to reduce total nodule mass.

- Somasegaran, P., Reyes, V. G., and H. J. Hoben, 1984. The Influence of High Temperatures on the Growth and Survival of *Rhizobium* spp. in Peat Inoculants During Preparation, Storage, and Distribution. Can. J. Micro. 30 (1): 23-30.

Gamma-irradiated peat was used to prepare inoculants for ten different species of tropical legumes and sent to cooperators in 14 cities in 13 tropical countries. Each cooperator received a package containing a maximum recording thermometer, plating media components, special instruction sheets and inoculants. Control experiments were pursued in the laboratory by exposing the various inoculants to 28, 37, and 46 degree C. Temperatures reached in the inoculants during their transportion varied from 26 degree C (Mexico) to 45 degree C (Kenya). Arrival time of the inoculants ranged from 6 days (Hissar, India) to 54 days (St. Augustine, West Indies). Although a total loss

of viability was reported for the chickpea inoculant (Saudia Arabia) and a severe decrease in two others (bean and lentil inoculants in Kenya and Saudi Arabia, respectively), over 90% of the inoculants received had viable counts in excess of 1 × 109 cells per g moist peat. Laboratory data indicated that 28 degree C was optimal for multiplication to maximal numbers in excess of 1 × 109 cells per g moist peat. Six inoculants studied for long-term storage showed excellent quality for 24 weeks at 28 degree C. At 37 degree C there was comparable cell multiplication as at 28 degree C with 8 of the 10 strains, only during the first week 46 degree C was lethal for all strains. Large reductions in viable counts, observed during inoculant preparation when broth cultures are added to peat, may be attributed to a dilution undergone by the broth culture in the peat.

- Somasegaran, P., Woolfenden, R., and J. Halliday, 1983. Suitability of Oven-dried Root Nodules for *Rhizobium* Strain Identification by Immunofluorescence and Agglutination. J. Appl. Bact. 55:53-261.

Legume root-nodules, dried at oven temperature (70 degrees C for 48 h) were suitable for *Rhizobium* strain identification by immunofluorescence and agglutination. The fluorescence of bacteroids of *R. japonicum*, *R. leguminosarum*, *R. meliloti*, *R. phaseoli*, and *Rhizobium* spp. from oven-dried nodules was the same as those from frozen, desiccated, or nodules dried at room temperature (28 degree C). Oven-dried nodules did not require further steaming for agglutination. Bacteroid agglutination gave 2-16 fold lower titres than those of the cultured cells. Fresh and oven-dried soybean rhizobia from a mixed inoculation gave exactly the same results when identified by immunofluorescence or agglutination.

Conference Presentations

- Beck, D. P. (Presented by R. J. Davis), 1983. Legume Inoculant Technology. Conference on Impact of Microbioloby in Tropical Agriculture. Kuala Lumpur, Malaysia.

- Davis, R. J., Halliday, J., and F. B. Cady, 1983. Poster Session. 5th Int'l. Symposium on Nitrogen Fixation, Noordwijkerhout, The Netherlands.

- Davis, R. J., 1984. Potentials and Limitations of Biological Nitrogen Fixation. ISSS Commission IV Conference on Soil Test Crop Response Correlation Studies, Dhaka, Bangladesh.

- Halliday, J., 1983. Biological Nitrogen Fixation: What's in it for Hawaii? Presented at Hawaii Fertilizers and Ornamentals Short Course, Kona, Hawaii.

- Kilkenny, M. R., and D. E. Welsch, 1982. An Economic Analysis of Quantitative Targets for BNF. Agron. Abstr. p. 212.

- Munns, D. N., 1983. Coping with Soil Acidity in Legume Symbiosis. 7th Australian Legume Nodulation Conference, Canberra, Australia.

- Rangarajan, M., Somasegaran, P., and H. J. Hoben, 1983. Occurrence of Different Serotypes of *Rhizobium meliloti* in Hawaiian Soils. 6th S. Conf. Microbial Inoculants, Bangalore, India.

- Rangarajan, M., Somasegaran, P., and H. J. Hoben, 1983. Variation in the Fluorescence of Bacteroids of *Rhizobium japonicum* and *Rhizobium* sp. (*Vigna unguiculata*) from Nodules of Homologous and Heterologous Hosts. 9th North American *Rhizobium* Conference, Ithaca, New York.

- Rangarajan, M., Somasegaran, P., Hoben, H. J., and J. Halliday, 1983. Nodule Occupancy by Different Strains of a Multistrain Rhizobial Inoculant in Two *Leucaena* Cultivars. 6th Southern Regional Conference on Microbial Inoculants, Bangalore, India.

- Roskoski, J., and T. Wood, 1983. Poster Session. 5th Int'l. Symposium on Nitrogen Fixation, Noordwijkerhout, The Netherlands.

- Singleton, P. W., and B. B. Bohlool, 1983. Poster Session. 5th Int'l. Symposium on Nitrogen Fixation, Noordwijkerhout, The Netherlands.

- Singleton, P. W., 1983. Poster Session. 7th Australian Legume Nodulation Conference, Canberra, Australia.

- Singleton, P. W., and B. B. Bohlool, 1983. The Effect of Salinity on Nodule Formation by Soybean. 9th North American *Rhizobium* Conference, Ithaca, New York.

- Singleton, P. W., Burton, J. C., Halliday, J., Davis, R. J., and F. Cady. 1983. Developing Cost Effective *Rhizobium* Technology for the Tropics and Subtropics. Int'l Symposium on Soybean in Tropical and Subtropical Cropping Systems, Tsukuba, Japan.

- Somasegaran, P., 1983. Two methods for Inoculant Production with Diluted Liquid Cultures of *Rhizobium* and Autoclaved Peat. 9th North American *Rhizobium* Conference, Ithaca, New York.

- Somasegaran, P., Reyes, V. G., and H. J. Hoben, 1983. The Influence of High Temperatures on the Growth and Survival of *Rhizobium* spp. in Peat Inoculants During Preparation, Storage and Distribution. 9th North American *Rhizobium* Conference, Ithaca, New York.

- Welsch, D. E., and M. R. Kilkenny, 1982. The Contribution of forages to Farm Earnings in Southeastern Minnesota. Agron. Abst. p. 130.

- Whitney, A. S., 1983. Contributions to the N Economy of Both Wheat and Maize Due to Legume Intercropping. Proceedings of Workshop on 'Nitrogen Dynamics for Tropical Cropping Systems', Rodale, Penn.

- Woolfenden, R. B., and B. B. Bohlool, 1983. Population Dynamics of *Rhizobium japonicum* and *Rhizobium leguminosarum* in Host and Non-host Rhizospheres. 9th North American *Rhizobium* Conference, Ithaca, New York.

APPENDIX II. NifTAL Publications (1975-1983)

Akyeampong, M., 1980. The Effect of Different Strains of Rhizobium and Phosphorus Levels in Mungbean *(Vigna radiata)* and Soybean *(Glycine max)*. M.S. Thesis, Univ. of Hi., 65 p.

Ayanaba, A., Asanuma, S., and D. N. Munns, 1981. An Agar Plate Method for Rapid Screening of *Rhizobium* for Tolerance to Aluminum. IITA Research Briefs 2 (4): 5-8.

Ayanaba, A., Asanuma, S., and D. N. Munns, 1983. An Agar Plate Method for Rapid Screening of *Rhizobium* for Tolerance to Acid-Aluminum Stress. Soil Sci. Soc. Am. J. 47:256-258.

Beck, D. P., and D. N. Munns, 1984. Effect of Calcium on the Phosphate Nutrition of *Rhizobium meliloti*. Soil Sci. Soc. Am. J. In Press.

Beck, D. P. and D. N. Munns, 1984. Nodulation of Legumes in a P-deficient Soil as Affected by P-efficiency of *Rhizobium*. Soil Sci. Soc. Am. J. In Press.

Beck, D. P., D. N. Munns, and K. G. Cassman, 1983. Phosphate Nutrition of *Rhizobium* spp. Appl. Environ. Microbiol. 47 (2): 278-282.

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Cassmann, K. G., 1979. The Phosphorus Nutrition of Two Grain Legumes as Affected by Mode of Nitrogen Nutrition. Ph.D. Thesis, Univ. of Hi., 95 p.

Cassman, K. G., Munns, D. N., and D. P. Beck, 1981. Phosphorus Nutrition of *Rhizobium japonicum*: Strain Differences in Phosphate Storage and Utilization. Soil Sci. Soc. of Amer. J. 45 (3): 517-520.

Cassman, K. G., Munns, D. N., and D. P. Beck, 1981. Growth of *Rhizobium* Strains at Low Concentrations of Phosphate. Soil Sci. Soc. of Amer. J. 45 (3): 520-523.

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Cassman, K. G., Whitney, A. S., and K. R. Stockinger, 1980. Root Growth and Dry Matter Distribution of Soybean as Affected by Phosphorus Stress, Nodulation, and Nitrogen Source. Crop Sci. 20 (2): 239-244.

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APPENDIX III. PLAN OF WORK FOR 1984, NIFTAL CONTRACT

OBJECTIVE 1 — Germplasm

Develop further the ecological model for responses by tropical legumes to inoculation with *Rhizobium*. (Singleton)

Conduct experiments using the split-root technique and tracers to explain host plant control of early events during infection and nodulation of legume roots. (Singleton)

Compare field methods for quantifying nitrogen fixation in tropical soils at different nitrogen and phosphorus levels. (Singleton)

Continue characterization of selected strains listed in the 1984 *Rhizobium* Catalogue for NifTAL's 50 target legumes. Strains will be characterized biochemically, for their intrinsic antibiotic resistance, for membership in cross-inoculation groups, and for growth in media of differing pH. (Somasegaran/Hoben/Halliday)

Continue revision and update of the *Rhizobium* Catalogue. (Somasegaran/Hoben)

Examine ecological traits of fast- and slow-growing soybean rhizobia, as they relate to previous cropping history, under field conditions in the People's Republic of China. (Dowdle)

OBJECTIVE 2 — Antisera

Prepare a catalogue listing antisera available from NifTAL, and write a manual on their production and use. (Hoben/Somasegaran)

Replentish antibody stocks for the most commonly requested types: *Phaseolus vulgaris*, *Glycine max*, *Vigna unguiculata*, *Leucaena leucocephala*, and *Cicer arietinum*. (Hoben)

Produce more FITC conjugated goat anti-rabbit globulin because of advantages offered by the indirect identification technique. (Hoben)

Develop antisera for strains added to the INLIT list to replace strains that cross react within the three-strain INLIT inoculant mixture (e.g. medic and pea strains), and strains for new legumes added to the INLIT list *(Phaseolus lunatus)*. (Hoben)

Compare serological reactions of bacteroids and cultured cells for each INLIT legume/*Rhizobium* combination. (Hoben)

Begin studies on effects of storage time and conditions on antisera quality. (Hoben)

OBJECTIVE 3 — International Network of Legume Inoculation Trials (INLIT)

Continue to offer INLIT A experiments on an extensive basis.

Follow-up with 20–30 effective cooperators on competition and persistence component.

An assessment will be made of the factors limiting completion of the trial program and submission of final harvest data.

Arrange meetings where possible in continental regions for coordination of INLIT implementation.

Issue an interim report of INLIT results.

Publish a paper on the INLIT methodology, analysis and interpretation of selected case studies as a HITAHR Bulletin.

Shift coordination of the Asian Network from Hawaii (Davis) to Bangkok (Beck) over the course of 1984.

OBJECTIVE 4 — Inoculant Development

Evaluate granular inoculants made from peat, charcoal, coal, and volcanic cinders for soybean, winged bean, and lima bean inoculation at field sites with chemical (low pH) and biological stress (high indigenous population of rhizobia). (Somasegaran/Hoben)

Publish the NifTAL Training Manual. (Somasegaran/Hoben/King)

Write and submit for publication a methods paper on the small, lab fermenter. (Somasegaran/Hoben)

Write and submit for publication a technical note on the simple transfer chamber. (Hoben)

Write and submit for publication a methods paper on production of research inoculants at NifTAL. (Somasegaran/Pinchin)

Write and submit for publication a paper on validation of the use of the dilution procedure for inoculant production with autoclaved peat. (Somasegaran)

Complete the Haleakala Inoculant Production Facility for development of low cost production systems, carrier testing, and for training inoculant producers from LDCs. (Somasegaran/Hoben/Burton)

Appraise the Inoculant Quality Incentive Program (IQIP) as an approach to ensuring that farmers worldwide can obtain quality inoculants. (Giovannini/Halliday/Burton)

OBJECTIVE 5 — Cropping Systems

Measure nitrogen fixation by tropical pasture legumes in a mixed pasture using isotopic nitrogen as per FAO/IAEA design. (Whitney)

Evaluate the use of isotopic nitrogen depleted material for assessing nitrogen fixation by *Leucaena leucocephala* and *Albizzia lebbek*. (Van Kessel)

Determine the effects of varying the density of corn and cowpea on nitrogen fixation by cowpea using isotopic nitrogen depleted tracer. (Van Kessel)

Compare isotopic nitrogen depletion with natural isotopic nitrogen abundance for measuring nitrogen fixation by crop and tree legumes. (Van Kessel)

Compare root growth and nitrogen fixation of grain legumes inoculated with rhizobia mixed with Azospirillum or Azotobacter. (Nakao)

Perform a *Phaseolus vulgaris* INLIT and two modified INLIT with *Phaseolus vulgaris* and corn (U.S. *vs.* Mexican cultivars). (Roskoski)

Determine the effects of tree *vs.* herbacious legume fallows on subsequent crop yields and nitrogen balance of the systems. (Roskoski)

Compare nitrogen fixation, nitrogen content of residues, mineralization of residues, and nitrogen uptake from residues of several grain legumes using isotopic nitrogen depleted material. (Roskoski)

Publish the proceedings of the Rodale Workshop on Nitrogen Cycling in Tropical Cropping Systems. (Roskoski)

Establish whether nitrogen can be transferred from a legume to an accompanying non-legume by mycorrhizae. (Hoben/Van Kessel/Halliday/Habte)

Examine the interaction of rhizobia and mycorrhizae in the rhizosphere. (Habte/Aziz)

Compare green manuring, crop rotation and intercropping with legumes for providing biologically fixed nitrogen to crops that are not legumes. (Chaudhary)

OBJECTIVE 6 — U.C. Davis Subcontract

Determine the tolerances for salt, acid, and phosphate in *Phaseolus lunatus*, *Lablab purpureus*, *Vicia faba*, and species of Acacia, Calliandra, Leucaena, and Sesbania.

Characterize mechanisms of acid and aluminum tolerance in rhizobia and symbiotic systems.

Relate tolerance of a strain to the conditions from which it was isolated.

Validate lab and greenhouse screening methods as indicators of stress tolerance in the field.

OBJECTIVE 7 — Univ. Minnesota Subcontract

The next phase of this activity involves an in-country study to test and appropriately modify the model to cope with parameters encountered in developing countries. Thailand, and specifically its Northeast region, has been selected for the field study.

OBJECTIVE 8 — BNF Resource Center

Conduct collaborative research to quantify nitrogen fixation under field conditions in developing countries.

Conduct collaborative research on competition and persistence of *Rhizobium* and in tropical soils.

Conduct collaborative research on soil and environmental stress effects on nitrogen fixation by legumes.

Conduct collaborative research on the interaction of rhizobial and mycorrhizal symbioses.

Offer internship/exchange opportunities at the Center for researchers from the region.

Reinforce the existing research network begun under the INLIT program.

Visit BNF researchers and relevant agencies to determine county-by-country needs for development support in the BNF area.

Respond to the extent possible to requests for development support on BNF from agencies/governments of the region.

Provide continuing technical information services, including a regional newsletter.

Collaborate as fully as possible with the programs of other agencies involved in agricultural development of the countries of the region.

Develop materials for promoting the improved use of BNF-based agrotechnologies.

Conduct an international workshop on *Azolla* as a nitrogen source for rice production systems.

Mount a BNF workshop for extension leaders in Bangladesh.

Mount a regional BNF workshop in Bangkok for FAO field officers. FAO confirmation of this request is pending.

Participate in design and delivery of an inoculant production workshop on Maui.

Offer one graduate assistantship for a candidate from the region to undertake PhD studies on nitrogen fixation by tropical agricultural legumes at a U.S. University.

Distribute training/extension materials as appropriate in the region.

Hold a meeting of the Center's Steering Committee.

OBJECTIVE 9 — Training

Mount a training workshop for extension workers in Bangladesh.

Mount a training workshop for agricultural extension leaders in Thailand.

Publish NifTAL/FAO pocket book for extension workers on legume inoculants and their use. Translate same into Spanish.

Mount a training course on commercial scale legume inoculant production.

Develop a manual on inoculant production.

Select host institute and prepare for Legume/*Rhizobium* Technology course to be held at an African Venue in 1985.

Sponsor 2 Intern Trainees (Peru and Ghana).

Host up to 10 Intern Trainees sponsored by other agencies. (Bangladesh and Sri Lanka)

Publish a general guide to the resource for development represented by NifTAL and cooperating institutions.

Publish and distribute the BNF Bulletin.

Computerize the NifTAL Document Collection.

Continue operation of the Visiting Scientists and Visiting Citizens programs.

A technology is available for establishing effective nitrogen-fixing symbioses between legumes and rhizobia. NifTAL continues to emphasize technologies to maximize the quantity of nitrogen fixed by legumes, e.g., rhizobial strain selection and seed inoculation.

APPENDIX IV. VISITOR PROGRAM

In the Visiting Scientist program, the aim is to meet informational needs as defined by the visitors themselves, as well as describing and demonstrating NifTAL's activities in the area of professional interest of each visitor. Visits last a day or even a few weeks. Longer visits are possible, but under the Intern Program (p. 10). NifTAL cooperates with USDA and other organizers of visits by foreign scientists to U.S. institutions.

Visiting scientists

John Vendeland	A & B Corp., HI
James Phukoa	A & B Corp., HI
Alan Higashi	A & B Corp., HI
Donn Munns	UC/Davis
Dick Green	Agron. & Soil Sci., UH
Hago M. AbdelMagid	Fac. of Ag., Sudan
Mitiku Habte	Agron. & Soil Sci., UH
Taufiqul Aziz	Agron. & Soil Sci., UH
Shiva Chaudhary	Agron. & Soil Sci., UH
Peter Rotar	Agron. & Soil Sci., UH
Frankie Sawyer	Auburn Univ., Alabama
Walter Herath	Winged B. Inst, Sri Lanka
Manibhai Desai	BAIF, India
D. S. Gorhe	BAIF, India
Rhonda Lincoln	Extension Serv. UH
Rick Koglin	MauiNet Proj, UH
Anson Bertrand	Office of Agri., S&T, USAID
Tejpal S. Gill	Office of Agri., S&T, USAID
Paul Moore	USDA, ARS, HSPA, HI
Robert V. Osgood	HSPA, Aiea, HI
Ruth Ann Taber	Plant Sciences, TX A&M U
Tran Phuoc Duong	Univ. of Cantho, Vietnam
Emil Tedeschi	Winery Owner, Maui
Haryono Semangun	LPP Yogyakarta, Indonesia
Anne Stewart Li	ABA International Inc.
K. Shyo	A&B Inc.
Misao Senaga	Ag. Student, Okinawa, Japan
Hiroki Ishikawa	Ag. Student, Okinawa, Japan
Keikan Uesato	Ag. Student, Okinawa, Japan
Ken Iha	Ag. Student, Okinawa, Japan
Tomohiko Teruya	Ag. Student, Okinawa, Japan
Seiichi Nohara	Ag. Student, Okinawa, Japan
Seigo Inoha	Ag. Student, Okinawa, Japan
Chetoku Iteshiki	Ag. Student, Okinawa, Japan
Yoshiyuki Takara	Ag. Student, Okinawa, Japan
Mutsuo Nakamoto	Ag. Student, Okinawa, Japan
Masumi Shingki	Ag. Student, Okinawa, Japan
Andrew Mantgki	HSPA, Aiea
Don M. Preivat	HHL 1-299 Infantry
Ernesto Ribucan	HHL 1-299 Infantry
John Musser	Gen. Services Foundation
Robert Musser	Gen. Services Foundation
Marcie Musser	Boulder, Colorado

W. C. Mitchell	Entomology Dept., UH
Warnel Decker	Univ. of Missouri
Colin Ramage	UH Manoa
Boyd Post	USDA, CSRS, Washington
Kent Kobayashi	Horticulture Dept., UH
W. G. Duncan	Univ. of Florida
Gregory Sullivan	Auburn University
H. Ikawa	Agronomy & Soils, UH
Duane Bartholomew	Agronomy & Soils, UH
Pacifico Mamasil	CMO, Mindanao, Philippines
Vicente Bargamento	Ceby, Philippines
Aung Khin	Ag. Research Inst., Burma
Khin Zaw	Ag. Research Div., Burma
Win Maung	Ag. Corp., Burma
Trillion Hmun	Ag. Research Inst., Burma
Colin Hughes	Commonwealth Forestry, Eng.
Rick Van Den Beldt	Agronomy Dept., UH
Marie Torres	Intec—Chile
Bob Shleser	Oceanic Institute, Oahu
Richard Nakahara	Facilities Planning, UH
Taka Izumi	CTAHR, UH
Charles Smith	USDA/CSRS, Washington
Orlando Almarza	Nat'l Weather Serv., Hilo
Goro Uehara	Agronomy & Soils, UH
Doug Godwin	IFDC, Arizona
H. Ikawa	Agronomy & Soils, UH
R. N. Roy	FAO, Rome, Italy
John Thompson	ICRISAT, India
Fred Beinroth	Univ. of Puerto Rico
Hari Eswaran	SMSS, Washington
Lafayette Young	Enegy Consultant, Maui
Tzo-Chuan Juang	FFTC/ASPAC, Taiwan, ROC
Tim & Karen Weiland	Allied Corp., Syracuse, NY
Wilson Kagabo	Monrovia, Liberia
Keith Archibald	Univ. of West Indies
Ahmed Orabi	Agri. Res. Cen., Egypt
Dennis Howell	Univ. of Guelph, Canada
A. Jean	McDonald Univ. of Guelph, Canada
Dr. Ahmed Osman	Soil Div., ACSAD, Syria
Bernie Carroll	Canberra, Australia
Jerry Bisson	Philippines, USAID
Michio Aragi	Kyushu Ag. Exp Stn, Japan

An awareness of the U.S. contribution to the third world through development assistance in agriculture is also promoted by a program under which NifTAL staff offer services to schools, clubs, citizen groups, and science fairs.

In the Visiting Citizen program, the objective is to increase awareness among local people (and our many tourists from the mainland) about the contribution being made by the United States to the development of agriculture in poor nations. A lay description of NifTAL's operations gives a glimpse of the technologies under development by NifTAL to solve agricultural production constraints in the third world.

Visiting Citizens

Naomi M. Z. K. Ariom
Iwalani, Keoni,
& Kawelo Darisay
Valerie K.
Alberta Alphen
Enid Olson
Martha Bissen
Vickie Kin Choy
Valery Pinheiro
Robert & Eleanor Lee
Chuck Davis
Ernest & Evelyn Thunell
Dave & Marilyn Reed
Vicky Foster
Karen Steinkirchner
Mark Redman
June Pastouil
B. Tanaka & Class
Rose Cabayacruz
Susie Dalumbar
Erlinda M. Salbigsal
Prudencia Tesoro
Carmelo Cordeiro
Alejandro Sarmiento
Tim Turiaga
Bert Najera
Lorito Lopez
Ray N. Saluatierra
Nathan D. Culim
Greg Albutra
Napoleon Bacayo
Lolito Reynon
Pedro P. Brinya
Mildred Hazama
Arthur, Dorothy & Robyn Reed
J. Delacruz & Class
F. Carter
E. Griffiths
Mr. & Mrs. E. J. Anderson
Ralph Moltzau
M. Nakamura
Alan Yamaguchi
Neil Swanson
Frances Sydow
Clark & Jean Nakamoto
Roy Nako
Ronald Richmond
John Mitchell
Ronald & Ann Guyton
Joann Tedeschi
Walter & Nora Fong
Isaac Stant
Warren Perreira
Cathy Kalehuawehe & Class
Gay Chang & Class
Frank Altz
June Alba
Joyce Miyahira
Maile Scanlan
Roy Cordeiro
Peter Banks
Tomthpamamal
Dick & Elnora Leyes
Bruce Loring
Margaret &
Frank Elizares
Takashi Takemoto
Darrel Lynch
Nasir A. Saidy
Mary Hanna
Kaye Ichiriu

Scientists of tomorrow... learning is fun at NifTAL with lots to see and do.

INTERNS (Visiting Scientists)

FUNDING SOURCE
S=NifTAL Shared
T=NifTAL Total

NAME	LENGTH OF STAY	COUNTRY	
H. AbdelMagid	6 months	Sudan	T
J. N. Chui	4 months	Kenya	S. (USAID)
M. Rangarajan	4 months	India	S (FAO)
S. (Seydou) Sanogho	2 months	Mali	T
U. Hla Than	4 months	Burma	S (FAO)

GRADUATE STUDENTS

NAME	PROGRAM	COUNTRY	
Taufiqul Aziz	Ph.D.	Malaysia	T
Shiva Chaudhary	Ph.D.	Nepal	T
Stephen Dowdle	Ph.D.	USA	T
E. Cuautle-Fabian	Ph.D.	Mexico	T

COSTA RICA COURSE

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