



**International Institute of
Tropical Agriculture**

Crop Improvement Division

**Activity Report
and
Work Plan
1992**

October 1992

Crop Improvement Division

Activity Report and Workplan 1992

Preface

The year 1992, IITA's 25th (Silver) Anniversary Year, concludes a quarter century of research to improve the quantity and stability of production of tropical Africa's most important food crops. During this year, for the first time, improvement activities for all the different crops have been grouped together administratively in the Crop Improvement Division. So it is timely to consolidate and summarize, all the Institute's crop improvement work in one document.

We intend to use this document in monitoring the direction, scope and progress of our work. A companion document, an archival annual report, will be produced by each Program or Unit after the year is completed, describing in detail the results of the year's work for each activity listed here.

This Activity Report and Workplan is organized by Program or Unit. Following an introduction, the work is described at two different levels: as (1) projects, and (2) activities (nested in projects). Each project begins with a rationale and background, including abstracts of a few key completed pieces of work. Current activities are then listed, with new ones (those starting in 1992) indicated by the symbol (§) in the title. To facilitate cross-referencing, each activity has a three-unit code of the form A.1.1: the first character is a letter to identify the Program or Unit leading the activity, followed by a number for the project and then a number for the activity within the project. The table of contents is a "summary of a summary" of IITA's crop improvement activities.

While mainly intended for internal use, this Activity Report and Workplan may also be of interest to others who want a comprehensive overview of what IITA is doing in the area of crop improvement. Additional copies can be obtained on request from the Director of Crop Improvement.

Mark D. Winslow
Interim Director

Crop Improvement Division
International Institute of Tropical Agriculture

List of Scientific Staff and Students

Office of the Director, CID

M.D. Winslow, Ph.D.

Interim Director, CID

Root and Tuber Improvement Program

R. Asiedu, Ph.D.

Leader, Breeder

A. Dixon, Ph.D.

Breeder

I. Ekanayake, Ph.D.

Crop Physiologist

K.V. Bai, Ph.D.

Cytogeneticist, consultant

M. Bokanga, Ph.D.

Biochemist

M. Porto, Ph.D.

Crop Physiologist, CIAT/IITA liaison scientist

S.Y.C. Ng, MSc.

Tissue Culture Specialist

Scientists in Special Project: ESARRN, Mozambique

M. N. Alvarez, Ph.D.

Breeder and network coordinator

J. A. Otoo, Ph.D.

Agronomist

Graduate Students

A. O. Akano

University of Ibadan, Ph.D. Nigeria

S. Akparobi

University of Ibadan, MSc. Nigeria

V. Boateng (Ms)

University of Ghana, MSc. Ghana

Y. Daraja

University of Ibadan, MSc. Nigeria

J. Ebiyau

University of Ibadan, MSc. Uganda

M.A. Fregene

University of Ibadan, Ph.D. Nigeria

R. Kapinga

University of Ibadan, Ph.D. Tanzania

M. Makame

University of Ibadan, Ph.D. Tanzania

V. Mbuyongha

University of Ibadan, Ph.D. Cameroon

P. Ntawuruhunga

University of Ibadan, Ph.D. Rwanda

C. Nwesigyne

University of Ibadan, MSc. Uganda

M. Simwambana

Univ. of West Indies, Ph.D. Zambia

N. Wanyera

University of Ibadan, Ph.D. Uganda

Maize Improvement Program

M. D. Winslow, Ph.D.

Leader, Breeder

S. K. Kim, Ph.D.

Breeder

J. G. Kling, Ph.D.

Breeder

J. M. Fajemisin, Ph.D.

Pathologist/breeder (joint with ICP)

M. Esseh-Yovo, Ph.D.

Breeder, Visiting Collaborative Scientist, Togo

N. Beninati, Ph.D.

Breeder (NCRE), Cameroon

Graduate Students

H. A. Akintoye

University of Ibadan, Ph.D. Nigeria

V. Adetimirin

University of Ibadan, Ph.D. Nigeria

Grain Legume Improvement Program

K.E. Dashiell, Ph.D.

Leader, Breeder

G.O. Myers, Ph.D.

Breeder

B.B. Singh, Ph.D.

Breeder and Officer-in-Charge, Kano Station

P.Q. Craufurd, Ph.D.

Breeder, Kano Station

I. Watanabe, Ph.D.

Physiologist (TARC), Kano Station (left early '92)

S.F. Blade, Ph.D.

Post-doctoral fellow

T. Terao, Ph.D.

Physiologist (TARC), Kano Station

S.M. Osho, MSc.

Food Technologist (IDRC)

R. Abaidoo, MSc.

Associate Expert (Niftal)

Graduate Students

M. Ogbaji

University of Ibadan, Ph.D., Nigeria

B. Kehinde

University of Ibadan, Ph.D., Nigeria

G. Ssemakula-Nankinga
I. C. Charles
P.M. Kormawa
N. N. Ntonifor
K. Mogotsi
H. Dapaah

Scientists in Special Projects

D.M. Naik, Ph.D.
R. Amable, Ph.D.
A. Doto, Ph.D.
A. M. Hossain, Ph.D.
J. Detongnon, Ph.D.

Plantain and Banana Improvement Program

D. R. Vuylsteke, PhD.
R. Ortiz, PhD.
S. Ferris, PhD.

Graduate Students

G. Harry
K.N. Mobumbo
B. Rubigiva
S. Witters

Genetic Resources Unit

N. Q. Ng, Ph.D.
S. Padulosi, Dott.

Graduate Students

F.N.U. Agwaranze,
A. Bakari,
S. I. Chukwuma,
C. P. Paul,

Biotechnology Research Unit

G. Thottappilly, Ph.D.
S. K. Hahn, Ph.D.
S. R. Schnapp, Ph.D.
H. D. Mignouna, Ph.D.
R. E. Ugborogho, Ph.D.
P. Petrilli, Ph.D

Graduate Students

A. M. Mih,
T. E. Njock
U. U. Ekuere

University of Ibadan, Ph.D., Uganda
University of Ibadan, Ph.D., Nigeria
Univ. of Hohenheim, Ph.D., Sierra Leone
University of Ibadan, Ph.D., Cameroon
University of Zimbabwe, Ph.D., Zimbabwe
Univ. of Guelph, Canada, Ph.D., Ghana

Leader/Breeder, (SADCC), Mozambique
Agronomist (SADCC), Mozambique
Breeder (SADCC), Mozambique
Breeder (GGDP), Ghana
Legume Specialist (NCRE), Cameroon

Leader, Tissue Culture Specialist
Breeder/Geneticist
Post-doctoral fellow, postharvest quality

Rivers State Univ. of Tech. Ph.D., Nigeria
Rivers State Univ. of Tech., Ph.D., Nigeria
Rivers State Univ. of Tech. Ph.D., Zaire
Program KUL, Belgium Ir. Belgium

Head, Genetic Resources Specialist
Plant explorer

Univ. of Ibadan, Ph.D., Nigeria
University of Ibadan, Ph.D., Nigeria
University of Ibadan, Ph.D., Nigeria
University of Ibadan, Ph.D., Sri Lanka

Head, Virologist. (on sabbatical leave)
Director Emeritus; Acting Head, BRU
Associate Scientist
Postdoctoral Fellow
Visiting Collaborative Scientist, Univ. of Lagos
Visiting Scientist, Univ. de Napoli, Italy)

University of Ibadan, Ph.D., Cameroon
University of Ibadan, Ph.D., Cameroon
University of Ibadan, Ph.D., Nigeria

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Tuber and Root Improvement Program

Cassava provides more than half of the dietary calories for over 200 million people in sub-Saharan Africa (about half of the total population). It also contributes substantial protein, through consumption of the leaves. It is Africa's food insurance: it gives stable yields even in the face of drought, low soil fertility and low intensity management. It can remain in the soil until needed, spreading out food supply over time, helping avert the tragic "boom and bust" cycle of oversupply followed by famine. However, diseases (African cassava mosaic virus, bacterial blight) insects (mealybug, grasshoppers) mites (green spider mite, red spider mite) and nematodes take their toll, occasionally in epidemic proportions. Postharvest processing is laborious: the high water content of the root must be reduced quickly to avoid spoilage and high transport costs, and the cyanogenic potential could pose a health risk if roots are not properly handled before consumption and if the nutritional status of a population is low.

IITA's founders invested in crop improvement on the premise that tropical food crops, having been neglected by research would probably show a large response to dedicated efforts in crop improvement. This hope was borne out in cassava, as much higher yielding, pest-resistant varieties (particularly to bacterial blight and mosaic virus) were quickly developed and rapidly adopted by farmers wherever planting material became available.

Despite the proven track record in cassava improvement, many challenges remain. High levels of green mite resistance are not yet available. Most of our work so far has been in the lowland humid tropics, but germplasm adapted to other agro-ecologies is also needed—particularly the cool midaltitudes and the dry savanna. Root dry matter and storability, cyanogenic potential and different flour qualities for various end-products need to be addressed. As always, increasing yield potential is a fundamental objective. The gene base for African cassava is uncomfortably narrow.

In contrast to the low-input cassava crop, yam is highly input-demanding (labor, agrochemicals)—yet still highly profitable because of its high market value. As a cash crop, it can help drive economic growth and employment. There are great opportunities for breeding to contribute to increased production and decreased input requirements. An erect (rather than spreading) shoot type might reduce or eliminate the high labor input for staking, and avoid some foliar diseases. Resistance to weevils, nematodes and foliar diseases could greatly stabilize production. However, progress so far has been quite limited because of recalcitrant flowering: controlled crossing has not been easy. Even if flowering were under the breeder's control, knowing which crosses might lead to viable hybrids would not be easy—phylogenetic relationships are little-understood, because chromosome numbers vary widely. If these problems could be overcome (or circumvented through biotechnology), it would create a new beginning for improvement of this crop.

T.1 Cassava germplasm introduction and evaluation

Project rationale

Cassava is not native to Africa—it was introduced from Latin America in the 1500's. Naturally, only a sample of the total gene pool was introduced; many species and ecotypes, representing the broader gene base of the crop, remained in the center of origin. It is thus important to accelerate the flow of germplasm from Latin America to Africa. Provision of new genetic diversity could bring in useful new traits and create new heterotic combinations that would produce higher yields. At the same time, research is needed to understand and identify which economically-useful traits might reside in this diverse germplasm base, and incorporated into African-adapted gene pools.

Activities

T.1.1 Broadening the germplasm base

M. Porto, N.Q. Ng and R. Asiedu

True cassava seeds are being obtained from CIAT headquarters in Cali, Colombia, taking into consideration parents' adaptation to different agroecologies. Four evaluation sites were identified in Nigeria, representing the humid, sub-humid, semi-arid and mid-altitude areas of Africa, homologous to those agroecologies where the parents were selected. A number of IITA cassava elite clones previously introduced to CIAT are also being used as sources of resistance to the African cassava mosaic virus (ACMV) in the introductions. National

programs in Africa are also contributing true seed populations to IITA which serve as useful sources of adaptation and quality characteristics. Other seed populations used to initiate the breeding process include: 1) progenies from open and controlled pollination of polyploid parents; 2) interspecific hybrids and their backcross derivatives; and 3) hybrids among IITA elite genotypes.

T.1.2 *In vitro* conservation and distribution

S.Y.C. Ng, R. Asiedu, A. Dixon, N.Q. Ng and J.B.A. Whyte

In addition to routine maintenance of the existing cassava and wild *Manihot* sp. germplasm used by the breeding program, 210 new accessions from Africa and Latin America were transferred from field collections to *in vitro* culture in 1991; over 100 accessions developed into plantlets. Maintenance of existing *in vitro* collections will continue as a major task in 1992, and more accessions will be transferred into *in vitro* culture. Regenerated clones will continue to be transplanted for virus indexing, certification and mass micropropagation for distribution and international testing. More advanced breeding lines will be cleaned up through meristem culture and heat treatment, confirmed virus-free by indexing and distributed to NARS. Twenty-four packages of cassava virus-free tissue culture materials were distributed in 1991 to 20 countries in Africa, Australia and United Kingdom. Improved germplasm in seed form is also distributed annually. Clone 80/40 in Sierra Leone and clone 30572A in Benin which are now being tested on-farm and multiplied for distribution to farmers originated this way. In 1991, 130 families of true cassava seeds of improved populations (210,000 seed) were distributed to 14 African countries for evaluation and selection under their local conditions.

T.1.3 Measuring diversity in cassava

A. Dixon, R. Asiedu, I. Ekanayake, Q. Ng, M. Bokanga, J.A. Whyte,
S. Jagtap, M. Porto, F. Schulthess, S. Yaninek and T. Ikotun

Biochemical markers for isozyme analysis are being established through polyacrylamide gel electrophoresis and isoelectric focusing as tools to assess genetic diversity in cassava. These biochemical fingerprints will be used in addition to morphological and physiological characteristics and pest and disease reactions to provide data for identification of landraces and improved germplasm, identification of heterotic groups and for hybrid confirmation, efficient selection of backcrosses derivatives, and investigation of phylogenetic relationships. These methodologies will also be useful to identify the spread of improved clones on-farm for impact assessment.

T.1.4 Cassava germplasm evaluation

A. Dixon, R. Asiedu, S. Y. C. Ng,
M. Porto, M. Bokanga and I. Ekanayake

The standard selection scheme for germplasm evaluation targeting the humid, sub-humid, mid-altitude and semi-arid agroecologies of Africa starts with a seedling nursery at one site within each agroecology in Nigeria. Selections from these nurseries are cloned and evaluated in successive trials—preliminary, advanced and uniform yield trials—selecting for performance with respect to ecological adaptation and quality of produce at each site. Evaluation for low cyanogenic potential is a high priority. At the uniform yield trial stage more than one location is used per agroecology. The diastatic activity and pasting property of cassava flour have already been identified as the determinant factors for the bread making ability of the flour, so screening for bread making ability is underway. So far many clones have reached the most advanced stage in the humid and sub-humid ecologies.

T.1.5 Cassava evaluation at the Mbalmayo Station (§)

A. Dixon and M. Bokanga

Morphological characterization and assessment of reaction to prevalent pests and diseases as well as root quality traits like mealiness and cyanogenic potential will be carried out at IITA's new humid forest Station at Mbalmayo, Cameroon. Entries for future multilocational trials will be identified. A collection of local clones at Njombe will be duplicated at Mbalmayo.

T.1.6 Field performance of virus-free cassava (§)

A. O. Akano, R. Asiedu, S. Y. Ng,
H. Rossel and G. Atiri (Univ. of Ibadan)

When new breeding clones outyield local germplasm, the question often arises as to whether this is due to the virus-free nature of the new material, as a result of *in vitro* cleanup. Pilot studies on small plots at Ibadan

many years ago during the main planting season did not support this hypothesis—they did not show any yield increase due to virus cleanup of planting material. This was presumed to be due to quick natural reinfection once the material was planted, due to the heavy ACMV pressure at Ibadan. Because of the pilot nature of that study, the issue is still being questioned, so it will be repeated with a larger number of widely-adopted genotypes and over three locations (forest, transition and savanna).

T.1.7 Genetic variation in cyanogenic enzymes (§)

M. Bokanga and B.L. Møller (RVAU, Denmark)

Previous research indicates that the enzyme linamarase plays an important role in the degradation of cyanogenic glucosides in cassava during processing. Cassava clones are being characterized for linamarase activity in the leaves and roots to assess cyanogenic potential. Recently, B.L. Møller of RVAU, Copenhagen has elucidated the metabolic pathway of the biosynthesis of linamarin and lotaustralin in cassava. His work on etiolated seedlings will be reconfirmed on field grown plants. The methods developed will be used to assess varietal differences in key enzyme activities and/or corresponding genes, and to find a molecular explanation of low cyanogenesis. These enzymes and/or genes could later be turned into biomolecular probes that would serve to identify very low-cyanide clones to be used as parents in the breeding program.

T.1.8 Screening for flexible harvest time (§)

A. Dixon, M. Bokanga, I. Ekanayake and T. Ikotun

COSCA results indicate that farmers desire cassava that can be harvested as early as 6 months after planting but at the same time want the flexibility of extended harvesting. Until now our breeding program has selected on a 12 month harvesting cycle. A postgraduate student will evaluate improved clones and parents at earlier harvest times for yield and for deterioration in the ground after 12 months.

T.1.9 Screening for drought tolerance (§)

I. J. Ekanayake, M. Porto and M. Bokanga

The savanna has potential for expansion of cassava cultivation, but existing germplasm was developed for the forest environment, where drought risk is less. Cassava is a relatively drought tolerant species, but prolonged drought will reduce yields. Field trials during the dry season at Ibadan and at the Minjibir Station (dry savanna) are planned in 1992 to study the effect of drought at different growth stages on growth and root yield. Wild *Manihot* species, cultivars and breeding lines will be compared. Detailed physiological studies will be conducted on some accessions to elucidate the mechanisms of tolerance to drought. Field screening will be done in association with the breeders to characterize several genotypes. Complementary studies in the area of stress physiology are being carried out in order to assess the feasibility of using differences in the fibrous root system of cassava as a selection criterion for drought tolerance. Preliminary studies initiated in 1990 showed large differences among the 10 genotypes tested.

T.1.10 Screening for cold tolerance (§)

I. J. Ekanayake, M. Porto and M. Bokanga

The midaltitudes could grow more cassava, but low temperatures may be a constraint. There is a dearth of knowledge on cold tolerance in cassava. The temperature regime of the midaltitudes will be simulated in controlled environments (using the Conviron at IITA), complemented by field studies in a midaltitude site (Vom, near Jos). The mechanisms of cold tolerance will be investigated, and screening protocols developed.

T.1.11 Cassava adaptation to inland valleys (§)

A. Dixon, I. Ekanayake, A. M. Izac,
M. Mahmoud, T. Ikotun and Entomologist

Cassava is commonly grown in inland valleys on residual moisture after rice. Several elite cassava clones and standard varieties will be evaluated on an inland valley slope at Ibadan and a better understanding of the major constraints of this agro-ecology will be sought by comparing the effects of different water management practices, mulching, moisture supply and soil temperature on crop performance. A detailed soil-plant-atmosphere continuum study will be done using soil hydrology and crop physiology techniques.

T.2 Enhancement of African-adapted germplasm pools

Utilizing new diversity effectively requires extensive intercrossing and selection. The wide range of target agroecologies, farming systems, utilization patterns and consumer preferences for cassava in sub-Saharan Africa dictates that a number of different recombining populations will have to be handled. Methodological studies are underway concerning breeding procedures to maximize the efficiency of the recombination and selection processes, particularly as regards interploidy crosses.

Activities

T.2.1 Agroecology-based population improvement

A. Dixon, I. Ekanayake, T. Ikotun and Entomologist

Recurrent selection steadily accumulates desirable gene combinations over time. Polygene accumulation also brings about more durable resistances that are difficult for pests and diseases to overcome. Recurrent populations are being developed and improved for the humid forest, forest transition/moist savanna, dry savanna and mid altitude agroecosystems. A total of 25 segregating families from diverse crosses are at various stages of evaluation for each of the agroecologies. For the humid forest, selection emphasizes tolerance to high soil acidity, low soil phosphorus and low solar radiation as well as resistance to cassava green mite (CGM), African cassava mosaic virus (ACMV), cassava bacterial blight (CBB) and cassava anthracnose disease (CAD). For the forest transition/moist savanna, soil constraints are fewer but selection for resistance to CGM, ACMV, CBB and CAD is still important. In the midaltitudes, initial vigor and rapid growth under cold air temperatures as well as resistance to CBB, ACMV and tip die-back take priority. For the dry savanna, types are sought which survive under severe water stress, show high initial vigor and growth rate, and good leaf retention (stay-green) under drought; and secondarily, resistance to pests and diseases. For all agroecosystems these desired traits are sought in a background of good agronomic and eating quality. Selected superior individuals or families from any cycle of selection are candidates for advanced testing and distribution to NARS.

T.2.2 Selfing and outcrossing in cassava

A. Dixon, R. Asiedu and C.A. Iglesias (CIAT)

Cassava is a highly heterozygous plant. Sib mating or selfing for one to a few generations should unmask deleterious recessive genes, and careful recombination can exploit both additive gene action and heterosis. Fourteen parental clones and 47 S_0 plants from a broad-based population were selfed in 1991 and 1,429 seeds from these selfings will be evaluated. S_1 progenies from 6 parental clones and S_2 progenies from three others from 1990 selfings were evaluated, selected, selfed and advanced to S_2 and S_3 in 1991. These progenies will be evaluated and selected lines will be hybridized in all possible pairs for further testing alongside the original parents. Genetic male sterility has been shown to facilitate efficient hybridization, and enhances hybrid production in several crops including cassava. Sources of genetic male sterility from introduced Latin American germplasm will be selfed. Male sterile progeny will be crossed to improved IITA clones and subsequently backcrossed to recover the adapted background of the IITA material together with a reasonable frequency of male sterility to facilitate hybridization.

T.2.3 Back-up source pops for specific characters

A. Dixon, M. Porto, R. Asiedu, M. Bokanga and Entomologist

Clones with specific characters identified from IITA-CIAT germplasm exchanges are hybridized with IITA improved clones and populations. Cyclic evaluation is conducted to recover these traits in populations with a background of adaptation to African agroecosystems. Selected individuals or families from each cycle of selection can also be used as sources of new cultivars or as breeding lines. In this way, a back-up source population for low cyanide and mealiness of the boiled tuberous root is being developed. A half-sib family selection scheme of 16 families is being employed in improving a back-up source population for higher levels of CGM/CM resistance.

T.2.4 Inheritance studies in cassava

A. Dixon and R. Asiedu

Crosses are being evaluated to clarify inheritance patterns of resistance to CGM, root mealiness, carotene content and inner skin color. This information will increase our efficiency in incorporating these traits into elite material.

T.2.5 Interspecific hybridization in cassava

R. Asiedu, K. V. Bai, S.K. Hahn and M. Fregene

Interspecific hybridization of African cassava clones with wild *Manihot* species introduced from Latin America introduces useful genes and helps elucidate phylogenetic relationships. Several wide crosses were evaluated in 1991; resistance to green mite and high protein content of tuberous roots were identified in some hybrid combinations. Cross compatibilities were established and numerous backcross progenies were generated for further evaluation. DNA studies of phylogenetic relationships are continuing (PhD thesis). Interspecific hybrids involving 22 pairwise combinations (including reciprocals) of nine wild *Manihot* species are undergoing evaluation in the field. 127 new crosses including reciprocals among wild *Manihot* species and several backcrosses between BC₂F₁ derivatives of interspecific hybridization and 16 improved clones were made and will be characterized. Cytological and biochemical/molecular analyses of the parents and progenies are providing interesting insights into the genetics of the genus and will be useful in genetic mapping.

T.2.6 Induction of polyploidy in cassava

K. V. Bai and S. K. Hahn

Polyploidy was induced in the diploid clones TMS 30001, TMS 42025, TMS 63397 and TMS 30555 using the cotton-wood plug method with 0.5% aqueous colchicine solution for 24 to 36 hours. Even higher ploidy is being sought by colchicine treatment of triploid and tetraploid clones. The objective is to find out the optimum ploidy level for cassava and develop useful polyploids for the breeding program for improvement of cassava at the tetraploid and triploid level.

T.2.7 Aneuploidy in cassava

K.V. Bai, S.Y.C. Ng and S.K. Hahn

Trisomics and monosomics are useful in locating the chromosomes that contain particular genes. Progenies of triploids and asynaptic clones of cassava will be screened cytologically with emphasis on isolation of aneuploids such as monosomics, nullisomics and trisomics. Later, through pachytene karyology studies the extra chromosomes of the aneuploids will be identified, with support from biochemical characterization. Information on megasporogenesis in cassava is very limited. This is important to understand the potential for recombination in some crosses. Preliminary studies on early stages of development of the female gametophyte showed normal formation of linear tetrads. These studies will be continued to gather information on later stages of development.

T.2.8 Cytogenetics of *Manihot* hybrids

K.V. Bai and R. Asiedu

Twelve of the 22 wild *Manihot* species available at IITA have been used to produce interspecific hybrids with cassava as well as hybrids between wild species. Chromosome synaptic behavior is being studied in these hybrids to gather information on genome homologies and cytogenetic architecture of these species. Preliminary observations indicate differences in the extent of chromosome pairing and chiasma frequencies. At A-I, an inversion bridge plus fragment was observed in some species hybrids. Detailed studies are continuing. Karyotype analysis at pachytene stage of meiosis will be continued using criteria such as chromomere pattern, heterochromatic regions, relative length, arm-ratio, telomeres or knobs, nucleolar chromosomes, etc; and idiograms will be prepared. Later, banding (or differential staining) techniques will also be tried for mitotic metaphase chromosomes. These will lead to identification of the individual chromosomes based on their morphological features which will in turn help to identify the extra chromosomes of aneuploids. The ploidy status of progenies from crosses between 2x and 4x, 4x and 4x as well as from open-pollinations of 4x is predicted initially using morphological criteria such as leaf shape, thickness and plant vigor. Later the ploidy is confirmed through cytological studies of PMCs (pollen mother cells) at metaphase-I (M-I) and anaphase-I (A-I) of meiosis. Chromosome pairing configurations and A-I disjunction and distribution of chromosomes and pollen fertility are recorded. Two aneuploids were isolated for the first time through cytological screening of the open-pollinated progenies of two triploid clones. Both the aneuploids are hyper-diploids, one with eight ($2n = 2x+8 = 44$) and the other with ten ($2n = 2x+10 = 46$) extra chromosomes. Cytogenetic behavior of these aneuploids is being studied.

T.2.9 Tetraploid population improvement

S. K. Hahn, A. Dixon and K. V. Bai

Agronomically promising tetraploids which arose from spontaneous sexual polyploidization will be intermated to recombine the desirable genes involved. Selected progeny will be recycled to improve the tetraploid population.

T.2.10 Yield vs. moisture in tubers

A. Dixon, R. Asiedu and J.A. Whyte

The stability and variation of yield, yield components and harvest index of improved cassava evaluated in multilocational trials for two years in Nigeria are being studied to identify the most efficient selection procedure to circumvent the inverse relationship between total tuber weight and dry matter content.

T.2.11 Flowering in cassava

M. Simwambana and I. Ekanayake

A PhD thesis activity is being conducted to explain what climatic, edaphic and physiological factors cause flowering to occur at Ubiaja but not at Ibadan for the shy-flowering genotypes TME 1 and TME 2. For related activity on yam, see T.5.1.

T.2.12 Mechanisms of CGM resistance

A. Dixon, I. Ekanayake, M. Porto and S. Yarinek

Fifty cassava clones derived from cycle 0 of the back-up population for high CGM resistance are being evaluated to identify different mechanisms of resistance. The goal is to diversify CGM resistance sources and pyramid different mechanisms to give higher levels of resistance in elite cassava populations. Physiological parameters such as plant water relations, stomatal behavior and other morphological and leaf tissue characteristics will be evaluated to better understand the relation of water stress to mite susceptibility. The influence of phytoseid activity on CGM damage will also be investigated.

T.3 Cassava food quality

Project rationale

Many of IITA's elite cassava clones have yields far exceeding the yield of local varieties, but adoption by farmers also depends on performance and acceptability of their leaves and tubers in the various postharvest systems and end-uses in Africa. Additionally it is important that NARS scientists be able to respond to inquiries from their governments and from consumers on issues of cyanide in cassava. An understanding of mechanisms of detoxification during the various modes of processing is necessary to make the right choice when introducing new cassava varieties in a particular area, and to design breeding schemes for reducing cyanogenic potential.

Completed studies

Bokanga, M. (unpubl.). Cyanogenic potential of cassava.

The range of variability in the cyanogenic potential of cassava clones from IITA improved germplasm, local germplasm and germplasm recently introduced from South America was studied. There is a wide variation across varieties, between and within plants for both roots and leaves. The correlation between the cyanogenic potential of leaves and roots is weak, suggesting that the probability of identifying a clone with low cyanogenic potential in the roots based on the picrate leaf test is very low. The observation of highly uneven distribution of the cyanogenic potential of cassava among different plant tissues and organs, and between plants of the same clone led to the development of a sampling procedure which increases reproducibility.

Bokanga, M. (unpubl.). Cyanogenic potential and taste of cassava roots.

A survey of 160 cassava clones (local and improved) indicates that cassava roots can be classified in 3 taste categories : sweet, bland and bitter. The mean cyanogenic potential of sweet and bland tasting varieties was lower than that of bitter tasting varieties. However, the cyanogenic potential of some sweet tasting varieties exceeded that of some bitter tasting ones. Thus, although the "high cyanide" varieties tend to taste bitter, there are other varieties that have a high cyanogenic potential yet do not have a bitter taste. This finding suggests

that the cyanogenic compounds are different from the factors that impart bitter taste, so breeders may be able to separate the two characters through genetic manipulation. Hence both sweet and bitter varieties (desired by different consumers) could possibly be developed that are low in cyanogenic potential. The free sugar content of cassava tuberous roots does not have a masking effect on their bitterness.

Bokanga, M. (unpubl.). Processing and cyanogenic potential of cassava leaves and roots.

Although the cyanogenic potential of cassava leaves is 5 to 20 times that of the roots, toxicity is usually not associated with the consumption of cassava leaves. Any process that disrupts the integrity of plant cells causes degradative enzymes to come into contact with cyanogenic compounds, stimulating the hydrolysis of cyanogenic glucosides and the release of cyanide. Pounding of leaves or grating cassava roots disrupts cells and so enhances this process of detoxification. "Retting" (rotting, or microbial degradation of cells) through short term storage also has this effect. Contrary to wide spread opinion, fermentation contributes very little to the elimination of cyanogens from cassava. The residual cyanogenic potential of gari fermented for one to four days is similar to that of non-fermented gari. In the absence of fermentation (cassava leaves are generally not fermented), the pH remains in the neutral range thus favoring the hydrolysis of cyanogenic glucosides and the decomposition of cyanohydrins to produce the volatile hydrocyanic acid which is rapidly lost during cooking.

Activities

T.3.1 Simplified linamarase preparation

M. Bokanga

A major constraint to the adoption of the best methodology for determination of cyanogenic potential in cassava is the high cost of the enzyme linamarase. Existing methods for preparing the enzyme require a refrigerated centrifuge which is usually not available in NARS. A method for the preparation of the enzyme that does not require a centrifuge will be developed.

T.3.2 Cassava processing and detoxification

M. Bokanga, S. Essers (WAU, Netherlands)
and H. Rosling (Uppsala Univ., Sweden)

Acetone cyanohydrin, the hydrolytic product of linamarin is the compound responsible for food intoxication attributed to cyanide in cassava. Destruction of linamarin by the endogenous enzyme linamarase (in those varieties containing much of it) thus detoxifies cassava. The enzymatic breakdown of linamarase, the stability and decomposition of cyanohydrins, and the effect of processing conditions on these pathways will be investigated to further understand the toxicity potential of cassava and its products.

T.3.3 Nutrient supply and cyanogenic potential (§)

M. Bokanga and J.H. Bradbury (Australian National Univ.)

In addition to the current work on cyanogenic potential under water stress, controlled experiments will be conducted in greenhouse using pots containing fixed amounts of nutrients to assess the effect of the nutrients on the cyanogenic potential.

T.3.4 Bitter compounds in cassava roots

M. Bokanga and J.H. Bradbury (Australian National Univ.)

Because bitter taste and high cyanogenic potential are often found associated in cassava, it is commonly assumed that the same compounds cause both phenomena. However, previous work (see completed studies) found some clones that have a high cyanogenic potential without the bitter taste, suggesting that different compounds may be involved. To resolve this issue, the bitter compounds of cassava will be extracted, purified and their chemical structure determined using mass spectrometry and NMR spectroscopy.

T.3.5 Evaluating cassava root and leaf quality (§)

M. Bokanga, F. Nweke, O. Tewe (Univ. of Ibadan), and A. Larbi (ILCA, Ibadan)

The analysis of cassava food samples and information collected during COSCA Phase III will permit the identification of food quality characteristics important for cassava consumers. It will then be possible to evaluate cassava breeding materials for the most important characteristics. Analysis of the COSCA collections will include morphological (size, shape, color, etc.), physical (texture) and biochemical parameters

(nutrient content, cyanogenic potential, shelf life, etc.) of both leaves and roots. Both human and livestock nutritional aspects will be investigated.

T.3.6 Cassava flour for bread making

M. Bokanga and J.A. Delcour (KUL, Belgium)

Various handling and processing conditions will be evaluated so as to derive the optimum conditions for producing cassava flour with maximum bread making potential.

T.4 Yam Improvement

Project rationale

IITA's mainstream yam improvement activity is modest, in accordance with the Strategic Plan of 1989-2000. The effort focuses on conserving and characterizing germplasm, identifying useful new clones and distributing these for testing to national programs. Focused, strategic studies to break some of the barriers in yam hybridization are also being addressed—see T.5.

Activities

T.4.1 Yam germplasm characterization (§)

R. Asiedu, I. Ekanayake, A. Dixon, M. Bokanga, N.Q. Ng, and NRCRI

Our yam collection includes accessions from several species. However these are difficult to distinguish visually. Distinct criteria for identification are needed. A total of 1000 accessions (including 64 from NRCRI, Nigeria) will be assessed for their agrobotanical, morphological, biochemical, physiological and food quality characteristics in 1992 through a tri-locational activity (Ibadan, Abuja, Uтуру) jointly run between TRIP, GRU and NRCRI. Tests for food quality traits will consist of texture evaluation after boiling, starch quality, protein content and enzymatic browning. See also T.5.1.

T.4.2 Yam germplasm evaluation

R. Asiedu, C. Akem and Entomologist

Families (154) from crosses involving 111 parents were sown at Ibadan in a seedling nursery in 1991, mainly to evaluate reaction to viruses, anthracnose and nematodes. Promising seedlings will be cloned for further evaluation and hybridization. 1,284 clones selected among families from the seedling nursery in 1990 were planted in 1991 and are being evaluated at Ibadan for disease and pest resistance, quality characteristics and other agronomic traits. 673 white yam and 578 water yam clones from the 1990 clonal evaluation were grown in a preliminary yield trial in 1991 and are being further tested for disease and pest resistance, quality characteristics, and yield. 132 clones selected from the 1990 preliminary yield evaluation were tested in an advanced yield trial in 1991 and are being further studied for disease and pest resistance, quality characteristics and yield. 72 clones of white yam and 31 clones of water yam from 1990 advanced yield trials are under test for 2 years at 3 sites within the forest and transition environments.

T.4.3 Yam germplasm distribution

S.Y.C. Ng, R. Asiedu, G. Thottappilly and N.Q. Ng

Routine micropropagation of virus-free clones will be continued during the year for distribution to NARS. Fifteen packages of virus-free yam plantlets and microtubers were distributed to 14 countries throughout the world in 1991. Micropropagation of six certified virus-free white yam clones was also carried out. An additional five selected white yam clones were successfully regenerated and cleaned of virus through meristem culture and heat treatment. In 1992 the regenerated clones will be transplanted for virus indexing, certification and micropropagation for distribution and international testing through NARS. More yam clones will be subjected to virus cleanup. Virus-free white yam plantlets of five selected clones were established in sterile soil in a vector-proof greenhouse in 1990. About 1,000 plantlets were planted. After eight months of growth, over 1,800 minitubers were harvested from 850 mature plants. The minitubers were distributed to NARS. This was repeated in June 1991 and the minitubers will be obtained in February 1992.

T.4.4 Plant age effect on yam meristem culture (§)

S.Y.C. Ng

This study will determine the best time during the growing season to sample yam meristems for *in vitro* culture. Clones of *D. rotundata* and *D. alata* will be used.

T.4.5 Sprouting of yam microtubers (§)

S.Y.C. Ng and I. Ekanayake

Understanding the sprouting behavior of microtubers is important since these are being used for international distribution of elite germplasm. Initially, a large quantity of microtubers will be produced. They will then be treated with chemicals and stored under different conditions. Number of days to first sprouting and percent sprouting will be observed.

T.4.6 Field performance of virus-free yams (§)

S.Y.C. Ng, H.W. Rossel and I. Ekanayake

Field performance of virus-free yam plantlets and virus-free microtubers produced from them in the screenhouse will be compared against conventional, field-raised minisetts (chosen for similar weight to the microtubers) at Ibadan to evaluate the effects of virus cleanup. Plantlets will be multiplied and transplanted for hardening before field-planting. Plant establishment rate, virus reinfection rate and tuber production will be measured.

T.5 Yam germplasm enhancement

Project rationale

Genetic variability in yam has been little-utilized because making crosses is difficult. Firstly, flowering is "shy" and asynchronous, and this seems to be strongly influenced by the environment. A second reason is that yam ploidy is complex and little-understood. Breeders need to know chromosome numbers, and understand genomic compatibility among yam accessions in order to plan and implement successful hybridization schemes. See also biotechnological approaches to this problem in B.2 and B.3.

Activities

T.5.1 Environment and flowering in yam (§)

I. Ekanayake and Mr. Okwor (NRCRI)

"Shy flowering" is a major constraint in yam breeding. Yams appear to flower more readily at Uturu in eastern Nigeria than at Ibadan where IITA breeding has been concentrated to date. To clarify this, a set of diverse accessions including some that do and some that do not flower at Ibadan will be planted at three locations: Uturu, Ibadan and Abuja. Weather data is available from Ibadan only. Days to flower, profusion of flowering, sex of flowers etc. will be measured to try to understand the mechanisms that differentiate location responses. This is part of a larger tri-locational experiment jointly run between TRIP, GRU and NRCRI described in T.4.2. For related activity on cassava, see T.2.11.

T.5.2 Cytogenetics of yam species & hybrids (§)

K.V. Bai, R. Asiedu, and S.K. Hahn

Meiotic events during microsporogenesis will be examined in leading or elite genotypes from each species of food yams important in Africa, and in hybrids between them. Observations will be made on the degree of pairing, chromosomal aberrations, dissimilarities among putative homologs, etc. These will help us understand the nature of potential difficulties breeders will face in making particular interspecific hybrids for yam improvement.

T.5.3 Chromosome numbers in the yam collection (§)

K.V. Bai, R. Asiedu, and S.K. Hahn

Somatic cells will be examined during mitotic metaphase to determine chromosome numbers in the IITA yam germplasm collection. This information base will be used by breeders to plan crosses in a more rational way.

T.5.4 Stomatal traits and ploidy in yam

R.E. Ugborogho, S.K. Hahn, N.Q. Ng

Several cultivars of eight species of yam (*D. alata*, *D. bulbifera*, *D. cayenensis*, *D. dumetorum*, *D. esculaenta*, *D. mangelotiana*, *D. praeheensis* and *D. rotundata*) are being investigated for 1) foliar stomatal distribution, types and sizes and 2) chromosome numbers. This information may give us an indirect technique to more easily estimate ploidy levels, based on stomatal characters.

T.6 NARS collaboration and training

Project rationale

Collaborative research schemes help identify regional research priorities, catalyze information exchange, preclude duplicative research efforts, and increase the transfers of technologies among partners. Making use of the comparative advantage of NARS to conduct certain types of research benefits both the NARS and IITA. Feedback on IITA technology from NARS is invaluable in the continuous refinement of our own research strategies. Training strengthens the research capacity of NARS.

Activities

T.6.1 International testing of cassava

A. Dixon, R. Asiedu, S.Y. C. Ng, J.B.A. Whyte,
M.N. Alvarez, J.A. Otoo and NARS

Improved cassava clones developed for various target agroecosystems are distributed to NARS as *in vitro* plantlets. Performance is evaluated in multilocal trials in collaboration with NARS. The performance of ten such clones was evaluated in 1991. Results of two years' testing indicate superior performance of TMS 30572, 4(2) 1425 in Ghana, Togo, Benin and Nigeria and TMS 50395 and TMS 91934 in Ghana and Nigeria. These have been recommended for on-farm testing and are being multiplied for distribution to farmers under the EEC/IITA-OFAR project. Another set of 19 clones established and multiplied in these NARS are completing their first year of testing in 1992. They will also undergo multilocal testing in Sierra Leone and Guinea Conakry in 1992. Massive supply of more elite clones in tissue culture form will be enacted upon request from NARS. Post-flask management techniques will be emphasized in 1992 to make it easier for NARS to establish *in vitro* germplasm shipments.

T.6.2 National cassava trials in Nigeria

A. Dixon and NRCRI staff

Under the auspices of the Nationally Coordinated Research Project (NCRP) on cassava, we work with the National Root Crop Research Institute (NRCRI) of Nigeria in the evaluation of the performance of improved cassava clones nationwide. Ten IITA improved clones are being tested alongside NRCRI improved clones in 16 diverse environments. Eight new clones have been nominated for the 1992 trials and are being multiplied for supply to NRCRI. Clone TMS 91934 has been nominated to the National Accelerated Food Production Program (NAFPP) for multiplication and minikit trials in 1992 (pre-release stage).

T.6.3 Tropical Root and Tuber Crops Bulletin

A. Dixon and H. Mutsaers

The first issue of the newly reconstituted, more scientific Tropical Root and Tuber Crops Bulletin was distributed to all root and tuber crops researchers and NARS in January, 1992. One more issue will be produced in 1992.

T.6.4 Germplasm delivery to NRCRI, Nigeria

S. Y. C. Ng and E. N.A. Mbanaso

In vitro germplasm collection of cocoyams and sweet potato will be progressively duplicated at the National Root Crop Research Institute (NRCRI), Umudike, Nigeria, over the next few years. Initially, 100 accessions of sweet potato germplasm were sent.

T.6.5 Graduate training

1. Effect of genotype, stem cutting characteristic and root behavior on establishment ability of cassava. J. Ebiyau (MSc. thesis) and R. Asiedu.
2. Morpho-physiological adaptation of cassava to the mid-altitude agroecology. S. O. Akpanobi (MSc. thesis) and I. Ekanayake. (§)
3. Effects of environmental factors on flowering of cassava (*Manihot esculenta* Crantz). M. Simwambana (Ph.D. thesis) and I. Ekanayake. See T.2.11.
4. The effect of drought on establishment, early growth of cassava: mulching and genotype variability. Y. Daraja (MSc. thesis) and I. Ekanayake. (§)
5. Intercropping of cassava (*Manihot esculenta*) and sweet potato in semi arid zone of Tanzania. R. Kapinga (Ph.D. thesis) and I. Ekanayake.
6. Dry matter accumulation and assessment of cassava. P. Ntawurhunga (MSc. thesis) and A. Dixon.
7. Effect of reciprocal grafting on cyanide content in cassava. M. Makame (Ph.D. thesis) and S.K. Hahn.
8. Spontaneous polyploids in cassava: identification, agronomic characteristics, response to biotechnology. V. Boateng (MSc. thesis) and S.K. Hahn. (§)
9. Evaluation of virus-free cassava clonal materials. A.O. Akano (Ph.D. thesis), R. Asiedu and S.Y.C. Ng. (§) See T.1.6.

T.6.6 Training courses

1. Root crops research and technology transfer. May-June 1992.
2. Rapid multiplication courses in Kenya and Tanzania.

T.7 ESARRN Network

Project rationale

The East and Southern Africa Root Crops Research Network (ESARRN) brings together cassava and sweet potato researchers in the region and links them to IITA and CIP for technical assistance. Network efforts focus on coordination, training, germplasm development, postharvest technology and integrated pest management. The current phase ends Sept. 30, 1992.

Completed studies

Alvarez, M.N. and S.K. Hahn (eds.). 1989. **Root crops and low-input agriculture**. Proc. Third Eastern and Southern African Regional Workshop on Root and Tuber Crops, Mzuzu, Malawi 7-11 Dec. 1987. IITA, Ibadan, Nigeria. 269 pp.

IITA, 1989. **Root crops research collaboration in Africa**. IITA Meeting Report Series 1989/3.

Alvarez, M.N. and R. Asiedu (eds.). 1990. **The role of root crops in regional food security and sustainable agriculture**. Proc. Fourth ESARRN Workshop, Mansa, Zambia 29 Oct.-2 Nov. 1990. IITA, Ibadan, Nigeria. 153 pp.

IITA, 1990. **Cassava for tropical Africa: A reference manual**. IITA, Ibadan, Nigeria. 176 pp. (English, French, Portuguese).

ISTRIC, 1992. **Proceedings of a Symposium of the International Society for Root and Tuber Crops (ISTRIC)**, 20-26 Oct. 1991, Accra, Ghana (In press).

Activities

T.7.1 Regional research coordination

M. Alvarez, J. A. Otoo and NARS

To effectively utilize limited root/tuber research resources in the East and Southern Africa region, research planning and evaluation is coordinated among the region's NARS. Forums used include: steering committee meetings, head of program meetings, root crops collaborators meeting, root crop symposia, scientist exchange visits, technical monitoring tours and annual research reviews. Information on research activities is disseminated through production of proceedings of the meetings, and a twice-yearly newsletter.

T.7.2 Training

M.N. Alvarez and J. A. Otoo

Inadequate trained manpower is a major constraint to root crops research in the region. In-country training for technicians, extension staff and farmers is being emphasized through on-farm trials and technology transfer efforts, particularly in the areas of rapid multiplication of planting material, socioeconomics, and plant protection. Two in-country courses on rapid multiplication techniques are planned for 1992, in Kenya (July) and Tanzania (March). In-service training at IITA, Ibadan will also be conducted to increase skills in research methodology, rapid multiplication of planting material, and postharvest technology.

T.7.3 Characterization and diagnosis

J. A. Otoo, M.N. Alvarez, F. Nweke and NARS

The absence of detailed baseline information on root and tuber crops in Eastern and Southern Africa hinders research planning and evaluation of progress. Surveys and field trials are being conducted to correct this situation. Additionally, the COSCA project is collecting valuable data in Malawi, Rwanda, Burundi, Zambia and Kenya. Malawi and Zambia have completed Phase I; the others are nearing completion. Efforts will be made in 1992 to complete the surveys and reports.

T.7.4 Broadening the germplasm base

M. Alvarez and J. A. Otoo

The Eastern and Southern Africa region encompasses diverse agroecosystems in accordance with its highly variable topography, rainfall and soils characteristics. A broader genetic base is needed for root and tuber crops so that an array of clones adapted to the different environments can be identified. TRIP-Ibadan has sent useful seed populations and tissue culture materials for this purpose; these are being evaluated. More will be imported, including materials from CIP, both in seed and tissue culture form. Tissue culture facilities in one location will be improved to more rapidly multiply plantlets. More crossing blocks will be established to increase the volume of hybridizations. National programs in Malawi, Rwanda, Uganda and Zambia are capable of generating improved seed populations. Materials generated by NARS will be distributed for evaluation.

T.7.5 Evaluation & distribution of germplasm

M. Alvarez, J. A. Otoo and NARS

Promising germplasm selections are evaluated for yield, disease and pest resistance and consumer acceptability. On-farm trials including economic analyses are being intensified, as is the multiplication of elite materials so identified. Some good selections identified from germplasm sent by TRIP-Ibadan are currently being multiplied for distribution to farmers.

T.7.6 Postharvest technology

M. Alvarez and J. A. Otoo

Root and tuber crops are less well-known in Eastern and Southern Africa as compared to Western and Central Africa. More efficient methods of processing and utilization of these crops need to be identified to make them appealing to more consumers, and attractive for increased production, including commercial scale. This should result in increased demand and hence incentive for farmers to increase production. Surveys are underway to characterize traditional processing and utilization methods and identify their constraints. Simple processing equipment is being imported from IITA-Ibadan for evaluation. A processing facility established in Malawi will be improved in 1992.

T.7.7 Technology transfer

M. Alvarez, J. A. Otoo and NARS

Improved root and tuber crop technologies will have no impact if they are not transferred to farmers. Multiplication and distribution of improved germplasm will receive emphasis in 1992. Additional multiplication sites will be established. More in-country training courses for farmers and extension staff will be organized in 1992. Field days, demonstrations, and preparation of extension documents will be done by NARS.

Maize Improvement Program

Maize is in high demand in Africa for human consumption as a starchy base in a wide variety of gruels, porridges and pastes, as well as a fresh vegetable ("green maize", or corn on the cob). Industrial demand (brewing, livestock feed, starch) is also growing. Often intercropped with cassava in the forest, its contrasting growth cycle increases the overall system's efficiency of nutrient capture and use, and smooths out peaks in the time course of labor input and food output for the enterprise. In the moist savanna sorghum/millet belt and in the midaltitudes, the nitrogen responsiveness of maize has stimulated a boom in its cultivation for cash-crop sale to distant urban markets as well as to diversify local diets. This dramatic expansion is having important "ripple" economic benefits throughout the savanna area.

Africa's predominantly smallscale, home labor-based farmers appreciate the low labor requirement of maize cultivation: spacing is wide, minimizing the work involved in sowing, and creating room for intercrops and for easy movement during weeding; its vigorous, rapid vertical growth makes it highly weed-competitive; stooping to harvest is unnecessary; and the cobs form an ideal storage unit as-is. They also value the crops' pest resistance: ears are protected from birds, rodents and insects by a husk; and open-pollination ensures genetic heterogeneity, allowing rapid gene reassortment in response to new pest challenges, thus preventing severe epidemics. Marketing of maize is also simple: the tough dry grains are easily handled, transported and stored during retailing.

In view of the importance of maize in African farming, IITA has mounted a maize improvement effort since 1972. It has scored many notable successes, such as the creation of open-pollinated germplasm yielding 30-100% more than local varieties under a range of smallscale farmer management systems; hybrid germplasm adding another 15-25% yield increment; the first practical, large-scale system for breeding resistance to maize streak virus—work which earned IITA the CGIAR's highest accolade, the King Baudouin Award, in 1986; high and stable resistance to rusts, blights and downy mildew; increasing resistance to striga; and the incorporation of all these traits into a range of maturity, agro-ecology and grain-type classes to fit the diverse needs of the continent's farmers. These accomplishments, and the wide spread of the materials on-farm were in large part achieved through collaboration with national research scientists. Many mechanisms were (and are) being used to strengthen NARS capacity to develop, utilize and transfer improved technologies to farmers.

M.1 Maize Improvement for the savanna

Project rationale

Maize cultivation in the moist savanna expanded dramatically over the past 20 years due to north-south road construction (better market access) and the availability of (often subsidized) fertilizers plus more responsive varieties that could exploit it, largely developed by IITA. High and stable yield potential to maximize profitability in this system remains a central priority for research. Pressure on governments to reduce fertilizer subsidies is increasing, though; this may drive production costs up and farmers may cut back on maize in favor of lower-input, lower-output crops, reducing overall regional food production. To avoid this decline, increased internal generation of N in maize cropping systems and improved nutrient recovery by maize must be achieved. Although improvements in cropping systems will be most important to attain these goals, breeding a maize plant that is more efficient in capturing soil N can complement this. Drought is also a major abiotic constraint in the savanna; resistance breeding can help. A priority biotic production constraint of the savanna which appears highly amenable to resistance breeding is the parasitic weed striga. Additional important pests (although less important than in the forest) against which breeding has proven highly effective include streak virus, lowland and highland rusts and blights, stalk rots and storage insect pests. All these resistances need to be combined in an adapted genetic background including different maturity durations and grain quality characteristics desired by different producers and consumers.

Completed studies

Mareck, J.H. (unpubl.). Creation of base populations for the "comprehensive breeding system" to accelerate the improvement of yield potential in IITA maize germplasm.

Private sector companies are interested in hybrid development while most public sector agencies work only on OP's. Because of its unique responsibilities, IITA works on both. The "comprehensive breeding system"

(Eberhart et. al. 1992) is a scheme for producing both hybrid and OP germplasm in a coordinated way. It was adopted by IITA in the late 1980's to streamline the breeding program by utilizing the same base populations for both OP and hybrid improvement, unlike the separate efforts that preceded this. Combining ability studies were first conducted to categorize the better varieties according to heterotic groups. The best materials were then combined to create base populations that would express heterosis when crossed across groups. Thus, for late-maturing germplasm, TZL Comp 3 was created by combining TZB-SR and Suwan 1-SR (both belong to the Caribbean flint-grained racial group); it is heterotic to TZL Comp 4, created from Pop 21-SR, Pop 43-DMRSR, TZPB-SR and DMR-LSRW (dent grains, Tuxapeño racial group). For early-maturing germplasm, TZE Comp 3 is derived from TZESR-W and DMR-ESRW; it is matched against TZE Comp 4, comprised from Pop 30-SR and Pop 49-SR.

Kim, S.K. (unpubl.). Combining ability of tropical maize germplasm.

Studies of the combining ability of four groups of tropical maize germplasm were conducted to identify heterotic combinations that could be exploited for hybrid breeding. The groups were: 1) tropical OP populations (10 white and 7 yellow), 2) tropical x temperate inbreds (5 tropical and 5 temperate), 3) tropical inbreds (11 white and 13 yellow lines), and 4) 4) and inbred x tester crosses (70 inbreds with 4 testers, including Mo17, B73, a Tuxapeño line, and a Caribbean flint line). These trials were conducted over a period of 2-3 years in both forest and savanna environments. G x E analyses are in progress, but preliminary observations are as follows: Among the white-grained OP populations, TZB x Tuxapeño showed the highest combining ability. General combining ability (GCA) effects were more important than specific combining ability (SCA) effects for yield and some other agronomic traits. GXE effects were not significant for the populations across four lowland sites, but for inbreds, tropical x temperate crosses were highest yielding in the savanna, but tropical x tropical crosses yielded the most in the forest.

Kling, J.G. (unpubl.) Nitrogen use efficiency of adapted OP varieties.

Nine elite OP varieties and one hybrid check were grown at three fertility levels (0, 60, and 120 kg/ha N) at four locations in Nigeria, representing the major lowland ecologies of the region. In order to understand the physiological mechanisms associated with NUE (grain produced per unit of applied fertilizer N), dry matter production and N content of plant tissues were determined at silking and at harvest. Effects of N stress on phenology and yield components were evaluated. There were large differences among varieties for yield, yield components, dry matter production and a number of morphological and physiological traits, but relative performance of varieties was similar across N levels. G x N interactions were observed for length of grain fill and leaf chlorophyll measurements, but no G x N interaction was detected for grain yield. Reductions in yield under N stress were largely due to decreased kernel number and ear number, while kernel weight was relatively unaffected. Harvest index and shelling percentage were reduced only under zero N application in the savanna. Prolificacy (more than one ear per plant) is associated with nitrogen use efficiency, and IITA research in the mid-1970's suggested that TZPB tended to be prolific. Following on this, S1 families from TZPB-SR were evaluated for prolificacy under varying N fertility for the past two years, but little genetic variation was observed.

Kling, J.G. (unpubl.). Heterotic patterns of tropical inbreds and OP varieties under varying N fertility.

An experiment was conducted to determine testcross performance of 30 tropical inbreds under high and low N fertility. Lines were evaluated in crosses with 6 testers (180 entries total) representing three major heterotic groups, permitting classification of inbreds according to heterotic pattern. Comparisons can be made of heterosis under high and low fertility, providing information about the benefits of growing hybrids compared to OP varieties under high vs. low nitrogen conditions. Results are not yet summarized. G x N interactions were found for yield, in contrast with earlier findings in smaller sets of only elite populations that there was little GxN interaction. These results suggest that it may be possible to breed genotypes specifically for N stress conditions. The best lines will be recombined to produce an N-stress tolerant synthetic.

Akintunde, A.Y., G.O. Obilgeson, and S.K. Kim. Estimates of nitrogen use efficiency of hybrids vs. OP's. Senior author's M.Sc. and Ph.D. theses, Dept. of Agronomy, University of Ibadan, Nigeria; and Nigerian J. Agron. 5(1), 1991.

N use efficiency (NUE, defined as grain produced per unit of applied fertilizer N) and N utilization efficiency (NUTE; grain produced per unit of N taken up by the plant) of hybrid varieties, in comparison to OP's were studied for a Ph.D. thesis. Trials were conducted with four maize varieties and five N levels (0, 60, 120, 180, and 240 kg N/ha) at four sites in Nigeria (Ikenne, Ibadan, Mokwa, Samaru) for two years. Four varieties were tested including one single-cross hybrid, one three-way cross hybrid, one double-cross hybrid and one OP variety. Under the same fertilizer regime, hybrids out-yielded OP's in the savanna, so logically their NUE and NUTE were higher; mean of 48 and 52, respectively, versus 39 and 48 for the OP's (units are kg of grain per kg N). GxN interaction was not important.

Kim, S.K. (unpubl.). Performance of hybrid vs. OP varieties at different N levels and plant densities. Results of comparative trials (14 hybrid plus 6 OP entries) grown at two nitrogen levels (60, 120 kg N/ha) and two population densities (5.3 and 6.6 plants per m²) for two years at 5 Nigerian sites (Ikenne, Ibadan, Mokwa, Samaru and Bagouda) over 1987-89 indicated that TZB, one of the most widely grown OP's showed significantly lower grain yield under low N than the hybrids 8321-18 and 8644-31, consistently across forest and savanna sites. By definition this means TZB is lower in nitrogen use efficiency (NUE = grain produced per unit of applied fertilizer N) than the hybrids. Mean NUE in the transition zone was 32 and that in the savanna was 46, suggesting that savanna maize production may be more N-efficient than elsewhere, although this data is from only a few sites. 60 kg/ha was adequate to reach the point of "diminishing returns" (i.e. yield potential) in the forest, but maize continued to respond to higher rates (120 kg) in the savanna, in consonance with the greater productive potential of that environment. Yield decreased at the higher population density in the forest due to lodging and barrenness, but remained stable in the savanna. Results will be useful for modeling studies. A second set of experiments compared 8 hybrids against 4 OP's at 4 nitrogen levels (0, 60, 120, 180) and 3 plant densities (3.3, 5.3 and 6.6 plants/m²) at four sites (Ikenne, Ife, Mokwa, Samaru) from 1989-90; data are still being analyzed.

Kim, S.K. (unpubl.). Formation of leaf-rolling resistant OP populations.

During a chance 10-day drought in 1986 first season, striking differences in leaf rolling were noticed in a 6-week old inbred line nursery. TZDR-SR was formed as a synthetic from the six inbreds exhibiting the least rolling (KU 1414, 5012, 9499, 9848, INV138, SC43). All reactions were reconfirmed in pot studies with differing watering regimes in a hot glasshouse. Later that year, four more inbreds (1787, T220, SC 213 and Mo 20w; the latter eventually was shown susceptible) were identified by the pot technique and field performance at Bagouda; they were introgressed, along with three drought-resistant OP populations: CIMMYT's Tuxapeño Sequia, Latente (a South American pool that rapidly rolls its leaves when drought starts) and SAFGRAD's Pool 16 DR. After three cycles of random mating, half-sib families were screened under drought in northern Nigeria in 1987 while screening for streak at Ibadan. In 1988, S1 testing was conducted in the north but drought did not materialize. Also that year, greenhouse tests showed it to be striga-susceptible, so resistant germplasm was introgressed: 9006, 8425-8 and 8322-13. In the 1989 wet season, half-sib isolations were planted at Bagouda and Minjibir; severe terminal drought occurred at both sites and 295 ears were selected. After selecting it was noticed that nearly all the selections had the cytoplasm of the original 6-line synthetic, rather than the populations added later. In 1989 dry season at Ibadan, 295 yellow and 179 white half-sib families were again grown under streak but with just 50% of normal irrigation, and selection practiced for short anthesis-silking interval (ASI) and streak resistance; full sibs were formed from the selected families. Also, two more striga-resistant lines were added: 1368-STR (into the white subpop) and 9450 STR into the yellow subpop. The populations were again screened under drought in northern Nigeria in 1990. Divergent selection was practiced (drought resistance/drought susceptibility) in order to later be able to assess response to selection. Experimental varieties were formed in late 1990 for both divergent directions for future testing.

Kim, S.K. and M.D. Winslow. 1991. Progress in breeding maize for striga tolerance/resistance at IITA. CIMMYT, Mexico: Proc. of the Fifth International Symposium on Parasitic Weeds, Nairobi, Kenya 24-30 June, 1991.

Following the identification of STR inbreds in 1983, additional progress was slowed by the lack of a uniform artificial infestation technique. By 1990, a reliable technique was well-established. Key improvements were: lowering of the nitrogen fertilizer rate to 60 kg/ha, increasing the striga seed rate to 3,000 germinable seeds per maize hill, planting on ridges rather than the flat, and infesting directly into the holes where the maize is planted rather than broadcasting or row-drilling. By 1992 MIP had bred 9 STR (striga resistant) hybrids and 8 STR OP varieties and synthetics. OP material was derived from topcrosses of the STR inbreds onto adapted IITA OP germplasm. Although improvement is still ongoing, currently available material is well adapted to the savanna and moderately striga resistant. The best STR hybrids are 9021-18 STR and 9022-13 STR (both white-grained) and 9044-31 STR and 9025-8 STR (both yellow-grained).

Kim, S.K. and A. Y. Akintunde. (unpubl.). Performance of STR hybrids across nitrogen levels under striga infestation.

Four maize hybrids (two STR and two susceptible) were tested for three years (1989-1991) at Mokwa with several levels of nitrogen (0, 30, 60, 90, 120, 150, and 180 kg/ha) under artificial infestation. Striga emergence and damage was reduced significantly by higher N rates (90 kg and above). Application of about 60 kg/ha N was determined to be optimum for striga screening purposes. STR hybrids showed both reduced striga emergence (confirming resistance, rather than tolerance), reduced foliar, stalk and ear damage symptoms and higher grain yields than the susceptible hybrids.

Kim, S.K. (unpubl.). Genetics of resistance in maize to *Striga hermonthica*.

Genetic control of STR in 10 maize inbred lines was studied using diallel crosses and some F₂'s, for five years (1985-90) under artificial infestation at Mokwa. A quick visual rating (1-9 scale) of damage to leaves, stalks and ears was used to estimate host plant response. Results suggest that resistance is polygenic and additive. Recurrent selection should therefore steadily increase striga resistance.

Kim, S.K. (unpubl.). Effect of *Striga hermonthica* on yield and agronomic traits in maize.

Experiments were conducted to estimate effects of striga infestation on grain and stover yields and yield components of maize. An average of 67 percent reduction in yield under striga attack was estimated from 8 trials under severe natural (1984-85) and artificial (1988-89) infestation (0.5 g striga seed drilled per 2.5 m row, supporting 11 maize plants). All yield components (ears per plant, kernel number and kernel weight) were affected by striga.

Kim, S.K. and A.Y. Akintunde (unpubl.). Reactions of maize, sorghum and millet to collections of *Striga hermonthica* from different locations.

Thirteen Nigerian collections of *Striga hermonthica* were used to infest 4 maize, 5 sorghum, and 4 millet cultivars in greenhouse pots at IITA. The most aggressive collection as estimated by a multi-factor ranking of host response was from Bakura (Sokoto State) followed by Galadima (Borno State) and Mokwa (Niger State). Resistant and tolerant cultivars of maize and sorghum showed no interactions with the collections whereas the two millet cultivars showed significant host x collection interaction.

Mareck, J.H. and J.G. Kling (unpubl.). Single-gene conversion of white-grained base populations to yellow grain color.

White grain is in highest demand in Africa, but important sectors also desire yellow. White-grained germplasm can be easily converted to yellow by backcrossing with a single gene. This saves the expense of maintaining separate white and yellow populations for each target. The yellow-grain conversion system was developed over the period 1983-90 and found effective. It is now generally used for both OP and inbred germplasm.

Activities

M.1.1 Breeding for high yield in the savanna

S.K. Kim, J.G. Kling, M. Esseh-Yovo,
C. Thè (IRA, Cameroon) and M. N'Kishama (PNM, Zaïre)

The "comprehensive breeding system" base populations TZL 3 and 4 (full-season) and TZE 3 and 4 (early maturing) now form the core of both our OP and hybrid improvement efforts for both savanna and forest ecologies (see "Completed studies"). See details of their improvement in M.2.1. Additionally and specifically for savanna OP improvement, selected S1 families for two high-yielding savanna materials, Pop 29-SR C1 (750 families) and TZUT-SR C5 (462 families) will be recombined in 1992 to initiate the next cycle of selection. Emphasis in selecting the families in 1991 was on ear rot resistance (screened at Ikenne), so the next cycle should be better for a key weakness of this germplasm. For the southern, wettest part of the savanna, an extra late-maturing population (135 days) is being developed so the crop will mature closer to the end of the rainy season to reduce storage problems. There has been a request from Nigeria to convert population 49-SR to yellow grain color to get an intermediate-maturing yellow dent, which will commence in 1992. Forty-nine hybrids selected in 1991 will be yield-tested in the southern, central and northern (driest) parts of the savanna (Mokwa/Abuja, Samaru/Funtua, and Bagauda) in 1992. Hybrid 9021-18 STR (a new version of 8321-18) is particularly promising, with high yield performance and stability across testing sites—it will be re-tested. Five new hybrids are being recommended for the Nigerian National Zonal Trials in 1992. For the past two years, new "inter-agroecosystem" (midaltitude x lowland) hybrids have been multilocally tested in both mid-altitude and lowland ecologies; an additional year of testing may be conducted if necessary when analysis of 1991 trials is complete, including some new combinations of this promising material. A fourth combining ability study of temperate x tropical crosses is underway using IITA tropical inbreds TZI 3 and TZI15 and temperate inbreds Mo17 and B73. The University of Hawaii and Iowa State University participated in testing these crosses in their contrasting environments. Topcrosses with Togolese lines will also be made by our visiting collaborative scientist to identify best combiners.

M.1.2 Breeding for nitrogen use efficiency

J. G. Kling, H. A. Akintoye and S. K. Kim

Feasibility studies have been ongoing since 1989 to determine whether genetic diversity exists within IITA's main populations for improving N use efficiency (NUE) and to understand the physiological mechanisms

associated with N stress tolerance. As part of this effort, a graduate student is investigating the effects of heterosis ("hybrid vigor") on nitrogen use efficiency. Single crosses, double crosses and F2 progeny derived from four inbred parents will be compared for the second time under varying N fertility levels (0, 70, 140, and 210 kg N/ha) at Ikenne, Mokwa and Samaru. Results should show whether heterosis per se is one mechanism of NUE. Key physiological traits being measured are: dry matter production (at biweekly intervals), leaf extension rate and anthesis-to-silking interval. Research elsewhere indicated that "prolific" varieties (more than one ear per plant) may be more nitrogen-efficient, presumably due to their greater "sink strength" (greater mobilization of assimilates to the more numerous ear shoots). Efforts are underway to introduce prolific genes from a variety of sources into regionally-adapted germplasm. S1 families from crosses of tropical x temperate prolific germplasm will be evaluated under low plant density in 1992 and selections will be made for prolificacy, streak resistance, standability, and ear rot resistance. Selected lines will be intercrossed to produce prolific composites. These will be compared with semiprolific populations from CIMMYT in 1993 under varying fertility levels to determine their usefulness.

M.1.3 Root growth and nitrogen use efficiency (§)

J. G. Kling, W. Horst and M. D. Winslow

Studies have shown that 30-70% of soil nitrate is never captured by the maize crop. A collaborative project (GTZ-funded) with Prof. Dr. W. Horst will be initiated, studying the ability of varieties with more vigorous, extensive root systems to capture more nitrogen from the soil. A postgraduate student with experience in techniques for studying root growth is expected to begin work in 1992 or 1993.

M.1.4 Breeding for drought resistance

J. Fajemisin, S. K. Kim, D. Hema and J. G. Kling

DR (drought resistance) has been a major objective of IITA breeders in SAFGRAD since 1980, leading to the development of early (95 days) and extra-early (80 days) maturing, DR germplasm. Experimental varieties from selected Pool 16 DR C3 full-sib families will be tested multilocally by IITA staff and SAFGRAD collaborators in drought-prone areas of Nigeria (Bagauda and Minjibir), Burkina Faso, Cote d'Ivoire and northern Cameroon. Full sibs will be generated from Pool 16 DR C4 under streak pressure. In 1991, crosses were made between Pool 16 DR and: Pool 16 Sequia, the early fraction of Tuxpeño Sequia and La Posta Sequia. These crosses will be evaluated in 1992 and the best will be introgressed into Pool 16 DR next year. DR Comp. Early generated from 8 DR local and improved cultivars was crossed with a streak resistance source in 1991 and will be recombined and selected under drought in 1992. Ten experimental varieties with drought resistance (DR) were formed in 1990 from TZDR-W and TZDR-Y populations and were tested in 1991; the second year of testing will be repeated in 1992. Analysis of data from a study of the genetic control of drought resistance using generation mean analysis of diallel crosses among DR and susceptible inbreds will be completed in 1992 (Ph.D. project). Seven of the F1 crosses included in this study have shown outstanding yield performance under drought conditions for three years in Nigeria and Burkina Faso, and will continue testing in 1992.

M.1.5 Resistance to high soil temperatures (§)

J. G. Kling, M. D. Winslow and S. K. Kim

Early season drought was reported by farmers in village interviews as a major constraint in the savanna, causing considerable stand losses (Dr. J. Smith, RCMD). Seedling death may be due to high temperatures as well as moisture stress. Tropically adapted varieties from IITA will be evaluated in the screenhouse for tolerance to high soil temperatures by planting seeds in flats immersed in hot water baths at various controlled temperatures.

M.1.6 Breeding for striga resistance

S. K. Kim, J. G. Kling, V. Adetimirin, C. Thè (IRA, Cameroon),
M. Esseh-Yovo (DRI, Togo), P.Y.K. Sallah (CRI, Ghana)
and L. Akanvou (IDESSA, Cote d'Ivoire)

Over 5,000 genotypes are screened annually in fields artificially infested with *Striga hermonthica*. Within Nigeria, IITA staff will screen for striga resistance at Mokwa, Abuja, Samaru, Minjibir and Jos in 1992. STR breeding trials may also be conducted collaboratively with NARS in Cameroon, Togo, Ghana and Cote d'Ivoire. Genetic uniformity and striga resistance (STR) of current germplasm will be upgraded and the best sources for further population improvement will be identified. Evaluation of full-sib families from TZE Composite 5 (early, white, STR) will be carried out. Following on previous studies that found STR to also be

effective against *S. asiatica* but not *S. aspera*, further trials are planned to evaluate the stability of STR across striga species. *S. asiatica* work will be done in collaboration with Togo and possibly at a new Nigerian site near Abeokuta. A graduate student's genetic study will continue to determine the genetic control of striga resistance using generation mean analysis of crosses of inbred lines. Requests for STR seed for testing have been received from researchers in other IITA Programs, national program scientists, PASCON (Pan-African Striga Control)/FAO network, and private seed companies; seed will be increased in 1992 to meet these needs.

M.1.7 Screening for resistance to *Striga aspera* (§)

S. K. Kim and J. G. Kling

Germplasm resistant to *Striga hermonthica* may not be resistant to *S. aspera*, another widespread species. However IITA has done little work on *S. aspera* in the past. Two *S. aspera*-infested sites were located in 1991: Jos (Nigerian Police Staff College) and Bagouda (ICRISAT Station). Screening trials will begin at the Jos site in 1992, particularly for midaltitude material while the Bagouda site will hopefully be functional by 1993.

M.1.8 Inheritance of striga resistance in OP's (§)

L. Akanvou (IDESSA), J. G. Kling and D. Berner

Genetic variances and heritability of resistance to *Striga hermonthica* will be determined in an OP population, beginning 1992 (Ph.D. thesis). Full-sib families within half-sib families will be generated and evaluated under striga infestation. Selected families will be recombined and evaluated the following year, permitting a comparison of realized heritability with estimates of heritability obtained from the experiment in the first year. This study will provide valuable information concerning the genetics of striga resistance and the rate and extent to which resistance can be increased using conventional population improvement methods.

M.1.9 Disease-resistant popcorn varieties (§)

S. K. Kim and J. G. Kling

Specialty products can provide farmers with opportunities to generate extra cash, increasing equity. Popcorn presents such an opportunity; it was a popular snack food before its importation was banned in Nigeria in 1985 to conserve foreign exchange. In the recent EEC/OFAR meeting at IITA, NARS scientists from seven countries in West Africa urged IITA to develop adapted popcorn varieties, which are not currently available in the region. The initial approach will be to screen popcorn germplasm that has already been crossed to streak resistance sources, to create an OP population. Characteristics that will be improved are disease resistance (MSV, rust, and ear rot), popping expansion, palatability and yield potential. The next step will be to extract inbred popcorn lines from this pop that could be combined to form a high-performance OP synthetic. If demand warrants it, this could later be extended to developing hybrids.

M.2 Maize Improvement for the forest

Project rationale

Maize is traditionally important in the forest, commonly intercropped with cassava to benefit from their complementary growth cycles, canopy architectures and nutrient requirements, and to diversify the food supply. Some is consumed as a fresh vegetable ("green maize", an important cash crop) early in the season while some is left to mature, then stored as dry grain for gradual consumption. The long rainy season and constant high humidity incites fungal diseases and stem/cob boring insects, and inhibits grain drying, leading to storage pest problems. Nutrient-poor acid soils and low insolation limit growth and yield. Breeding in this harsh environment has nonetheless been successful in increasing on-farm yields by 30-100%. Resistance breeding has proven to be a highly effective approach for the control of fungi and viruses, and is showing promise against stem borers. The outlook for increasing storage pest resistance is also bright.

Completed studies

Kim, S.K. (unpubl.). Hybrids vs OP's across environments.

Hybrid and OP germplasm were compared (14 vs. 6 varieties respectively) across five sites spanning both forest and savanna in Nigeria; results are being analyzed. Preliminary results indicate that high plant density (6.6/m²) reduces yield in the forest, but increases it in the savanna. Diseases, insects and low radiation

appear to be the major causes of low yields in the forest.

Bosque-Pérez, N.A. and J.H. Mareck. 1991. Methods to screen for resistance to the maize stem borers *Eldana saccharina* and *Sesamia calamistis*. In Proc. Joint SAFGRAD Research Networks Workshop, March 7-14, 1991, Niamey.

Screening techniques to identify resistance to the maize stem borers *Eldana saccharina* and *Sesamia calamistis* are being continually refined. Screening against *S. calamistis* is done by infesting 21-day-old plants with 25-30 eggs (black head stage) obtained from a laboratory colony. Eggs are inserted between the leaf sheath and stem at the base of the plant, where they soon hatch and begin feeding. Damage ratings are taken at 2 and 4 weeks after infestation. Spreader rows are used to inexpensively amplify insect numbers while reducing the labor of infestation; it gave good results in 1990 and 1991. Strips of a susceptible maize variety (spreader rows) are planted one month prior to planting the test materials. Each strip consists of two rows (75 cm apart); a 7 m gap is left between strips. The test materials are planted between the strips and perpendicular to them, using 3 m row length and 1 m central alley. Spreader rows are artificially infested at silking with *Eldana* egg masses obtained from a laboratory colony. Adults emerge from the spreader rows and fly to the test plants to lay eggs. At maturity, ratings are taken on stalk breakage and ear damage.

Mareck, J.H. and N. A. Bosque-Pérez (unpubl.). Screening of diverse germplasm for resistance to *Eldana saccharina* and *Sesamia calamistis*, and the formation of breeding populations for stem borer resistance.

Inbred lines resistant (TZi 4) and susceptible (TZi 19 and TZi 28) to *Sesamia calamistis* were identified under natural field infestations at Umudike, Nigeria the early 1980's and have since been used as checks. Since the mid-1980's, the BR population of IITA (developed by screening for *Sesamia* at Umudike), the CIMMYT MBR (multiple borer resistant) population, MIR inbreds (maize inbred resistant lines from Hawaii), and a wide range of germplasm from North and South America which had shown resistance to other borer species were screened at IITA under artificial infestation. TZBR *Sesamia*-1 was formed in 1987 using 6 sources of resistance (CM 116, INV 575, Cateto Assis Brazil RGS x IV, Cateto Grande Mil, Costeno Mag. 350, and Cubano Cateto Ecuador 339) crossed to TZi 4. TZBR *Sesamia*-2 was formed in 1988 from TZi lines which had shown resistance in earlier screening (TZi 4, 6, 8, 9, 10, 17, and 18), but this population was discontinued in 1990 because it was found to be susceptible. TZBR *Sesamia*-3 was formed in 1988 from 29 lines, mostly from the CIMMYT MBR population, crossed to TZi 4. In 1985, 102 putatively stem-borer resistant introductions from the USA, CIMMYT, and ICIPE were testcrossed onto the hybrid 8338-1 to improve their adaptation and screened for resistance to *Eldana saccharina*. Significant differences were found for most assessments of *Eldana* damage. Following 2 more years of testing, 14 superior testcrosses were ultimately selected, and backcrossed to their introduced parent. This formed the basis of the TZBR *Eldana*-1 population. Inbred lines with tropical adaptation were screened for *Eldana* resistance in 1987 and the best 5 (TZi 2, 10, 12, 15, and ICAL 27) were recombined to form a synthetic, TZBR *Eldana*-2. Tropical, late-maturing, OP populations were also screened for resistance in 1987 and three populations, DMR-LSRW, La Posta, and TZSR-W-1 were selected for further testing. S1 lines from these populations were screened for *Eldana* resistance and selected lines were used to form TZBR *Eldana*-3.

Mareck, J.H., N. A. Bosque-Pérez and J. G. Kling (unpubl.). Evaluation of progress from selection for resistance to *Eldana saccharina* in three breeding populations.

Cycles of selection advanced under artificial infestation with *Eldana* were evaluated in 1989 (Cycles 0, 1, and 2 of TZBR *Eldana*-1) and 1990 (Cycles 0, 1, 2, and 3), confirming that resistance had indeed improved; ear damage and stalk breakage were progressively less in each cycle. By Cycle 3, ear aspect and grain yield exceeded that of the susceptible hybrid check, 8338-1. Plant height and maturity have inadvertently increased, however. This must be corrected in future selection since the population is intended for late sowings, when borers are most problematic; late sown crops need to have intermediate or short growth duration to mature before the rains cease. Cycles 0-4 of TZBR *Eldana*-1 and cycles 0, 1, and 2 of TZBR *Eldana*-2 were evaluated in 1991; results are still being analyzed.

Kim, S.K. and H. Rossel (unpubl.). The genetics of the "near immune" and "recovery" types of resistance to maize streak virus.

Different symptoms have been observed in different germplasms resistant to streak virus. A previous genetic study (Kim et. al., 1989, Crop Sci., 29:890-894) found that IB 32 (an inbred heavily used in IITA's resistance breeding project), which shows delayed onset of minor streak symptoms, was under oligogenic control and mainly additive. Following on this, two more types of resistance were examined: a) "near immune" (trace symptoms only) resistance in TZMi 301 and Population 10; and b) "recovery" (considerable yellowing in early growth stages but plant grows out of it) in TZi 15 and TZi 18. Generation mean analysis was used to study

resistance using F1, F2, and both backcross generations under artificial streak infestation. Type a) was found to be controlled by a single partially-dominant gene amenable to backcrossing. Type b) was polygenic, so it would require a recurrent selection process rather than backcrossing to transfer it in crosses.

Kossou, D.K., J.H. Mareck and N. A. Bosque-Pérez. 1991. Effect of husk cover on postharvest performance of improved and local maize varieties. In Proc. Joint SAFGRAD Research Networks Workshop, March 7-14, 1991, Niamey.

Two improved, high yielding varieties (Sekou 85 TZSR-W-1 and EV8443-SR), one local, improved variety (NH2) and one local unimproved variety ('Gbogbe' from Benin) were compared for resistance to *Sitophilus zeamais* in three storage forms: a) as husked cobs, i.e. with husks retained covering the cob- the predominant on-farm system; b) as dehusked cobs, and c) as shelled grain. Number of husk leaves was similar in all varieties (10 to 12) but the local variety Gbogbe had the longest husk extension (av. 9.5 cm beyond cob tip) and the fewest F1 weevils when stored as husked cobs under artificial infestation. Weevils took longer to develop in husked and dehusked cobs than in shelled grain, indicating that on-cob storage as practiced by farmers helps them resist weevil damage, particularly if husks are left on; and varieties differed in rank across these treatments, indicating there are genetic components of resistance associated with the assemblage of grains and husks onto a cob (see below). Comparisons of no-choice vs. free-choice showed no variety x treatment interaction, indicating that selection for resistance may be carried out using simple free-choice infestation methods.

Kossou, D.K., N.A. Bosque-Pérez and J.H. Mareck. 1992. Effects of shelling maize cobs on the oviposition and development of *Sitophilus zeamais* (Motschulsky). J. Stored Prod. Res. 28:187-192. Oviposition and development of the maize weevil, *Sitophilus zeamais* Motschulsky were studied on shelled maize kernels and unshelled cobs of the variety 'Blanc Deux Précoce' from the Republic of Benin. While weevils prefer to feed at the base of the kernel, on unshelled cobs they must feed and oviposit mostly on the kernel's crown, because grain packing restricts access to the basal portion. The resistance of unshelled kernels to the maize weevil was thus reflected in three phenomena: 1) reduced oviposition, as a result of non-preference of the crown as an egg-laying site; 2) longer weevil median development period (MDP) due to the less suitable endosperm diet near the crown; and 3) delayed emergence from the grain due to difficulty in finding a site on the kernel where the F₁ generation adult can emerge. These phenomena may explain the lower levels of weevil infestation found in traditional on-the-cob maize storage.

Kossou, D.K. and S. K. Kim. (unpubl.). Genetics of resistance to *Sitophilus* weevils.

Twenty hybrids, 6 OP varieties, and two sets of diallel crosses (12 x 12 white-grained and 13 x 13 yellow-grained) were tested as shelled grain under artificial infestation with weevils. Significant genetic differences were observed. Genetic analysis revealed non-additive gene action. Four of the best five yellow crosses had one common parent, TZi 30. An earlier study (1985, IITA Research Highlights) also reported differences among IITA germplasm for weevil resistance. The best hybrid identified at that time, 8329-15, also has TZi 30 as a parent. Although the relation to grain hardness was not specifically investigated, it is noteworthy that TZi 30 is dent-flint, i.e. softer than several flint parents in the diallel, suggesting that grain hardness differences would not fully explain differences in resistance.

Akinpelu, M., S.K. Kim and M.E. Aken'Ova (unpubl.). Genetic control of seed viability and vigor retention in stored maize grain.

Two inbred lines, 1368 and 5012 were found to differ in seed viability after a period of storage (high and low viability retention, respectively). They were crossed and F₁, F₂ and backcrosses were produced. Generation mean analysis is being used to elucidate the genetic control of viability retention. An index including rootlet and shootlet vigor was constructed to incorporate early "vigor retention" as well as viability *per se* into the evaluation. Data are being analyzed.

Mareck, J.H. and J. G. Kilng. (unpubl.). Conversion of dent and flint varieties to floury endosperm by single-gene backcrossing.

Maize varieties originally introduced into coastal West/Central Africa (forest zone) 500 years ago from Brazil/Central America by Portuguese traders were floury-grained types, so coastal African consumers naturally developed processing methods and a preference for this grain texture. Much high-yielding germplasm, however has dent grain type which is less preferred by traditional consumers. Floury grain was found to be a simple recessive trait that could be transferred by backcrossing to make varieties more appealing to coastal consumers. This scheme was used to make floury versions of the popular TZB-SR and TZESR-W. However some detrimental traits such as low yield were found associated with the gene transfer.

Activities

M.2.1 Breeding for high yield in the forest

J.G. Kling, S.K. Kim,

C. Thè (IRA, Cameroon) and M. N'Kishama (PNM, Zaïre)

Field observations in 1991 indicated that the reciprocal composite populations TZL Comp 3 and 4 (late-maturing) and TZE Comp 3 and 4 (early maturing), developed for the "comprehensive breeding system", have high yield potential in both forest and savanna environments. Until now, reciprocal S1 testcrossing has been used to improve them, but beginning in 1992 an S2 testcross system will be adopted to hopefully accelerate their improvement. Remnant seed of selected lines from both the early and late composite testcross trials have been sent to Cameroon. a) TZL: To maintain inter-population heterosis, a total of 169 testcrosses of TZL Comp. 3 C0 S1 families with TZL Comp. 4 were evaluated at two locations in Nigeria in 1991. Additional sets of this trial were sent to Cameroon and Zaire, at the request of NARS breeders. Based on this and other data, the next cycle of TZL Comp. 3 will be generated in 1992. S1 families from TZL Comp. 4 have been prescreened for resistance to maize streak virus, and will be evaluated for reaction to foliar diseases, downy mildew, ear and stalk rot resistance and other traits in replicated field trials in 1992. b) TZE: Similarly, 200 testcrosses of TZE Comp. 4 C1 S1 families with TZE Comp. 3 were tested at four locations in Nigeria as well as Cameroon and Zaire. Over 2000 S1's from TZE Comp. 3 C1 were evaluated at Ikenne for foliar diseases, ear and stalk rot, and other traits. A new cycle of TZE Comp. 4 will be formed in 1992. Testcrosses will be made from selected S1's from TZE Comp. 3 for testing in 1993. Hybrids: field superiority of hybrid maize over OP has not been clearly expressed in the forest environment over the years, so selection is continuing to find better material. Twenty-one promising hybrids were selected at Ikenne in 1991; yield testing of these and hybrids selected for downy mildew (38 hybrids), *Eidana* (24), and *Sesamia* (6) resistance will be carried out at the same site in 1992.

M.2.2 Breeding extra-early forest materials

J.M. Fajemisin and J.G. Kling

Green maize is an important revenue source for forest-zone farmers. The price is much higher the earlier it is harvested. To exploit this opportunity, extra-early varieties developed by SAFGRAD in the dry savanna to escape drought (60 days to green maize harvest) will be upgraded to resist forest diseases. TZEE-SR-W and TZEE-SR-Y will be advanced to BC 5 for streak resistance and also selected for resistance to blight, rust and curvularia.

M.2.3 Resistance to downy mildew

J.G. Kling, S.K. Kim, K.F. Cardwell and M. Omedeji (IAR&T)

Downy mildew (DM) is devastating on late-sown crops in parts of Nigeria and Zaire, and is spreading rapidly through the forest zone. Genetic sources of high resistance (DMR) are available but improved screening methods are needed in order to purify it in breeding populations. Zero susceptibility is probably not achievable, but we would like to aim for 10% or less in all OP populations. Streak-resistant conversions of the original DMR varieties from Thailand (Suwan 1 and Suwan-2) showed about 10% susceptible plants under artificial infestation in Akure in 1991. DMR populations developed jointly by NCRI/IITA (DMR-ESRW, DMR-ESRY, DMR-LSRW, and DMR-LSRY) had 15-20% susceptible plants. Upgrading is also needed in a number of CIMMYT populations and IITA composites. With the exception of TZE Comp. 4, the other major breeding populations (TZE Comp 3, TZL Comp 3, and TZL Comp. 4) already contain genes for DMR, but need upgrading. The best lines from TZE Comp. 4-DMRSR (a downy mildew version of TZE Comp. 4) will be backcrossed to TZE Comp. 4 C2 in 1992. To improve our screening accuracy in order to upgrade DMR in all these populations, several new techniques will be investigated in 1992, including a controlled environment (dew chamber) seedling infection system, and a daytime field inoculation method (which may reduce costs and mistakes associated with night work in the current method). Meanwhile, resistance levels in available DMR varieties will be monitored for the third time in 1992. Seed multiplication of these OP varieties will continue in anticipation of increased requests for resistant varieties from national seed production agencies. Among IITA hybrids, only three (8644-27, 8644-31, and 8444-32) have a resistant inbred parent (Ku1414-SR); the other parent is susceptible, so resistance of the hybrid is intermediate. Ninety-two new DMR hybrids were tested in Akure in 1991; five having resistance from both parental inbreds were selected and seeds are being multiplied. DMR conversions of IITA's most useful inbreds is underway; twenty streak resistant TZi lines are presently at the backcross 4 stage and will be screened in Akure in 1992. In addition to Ku1414-SR, 16 more resistant inbreds from Thailand have been introduced and are undergoing streak resistance conversion.

Several new DMR hybrids have been nominated for Nigerian National Zonal Trials in 1992, and Pioneer Hi-Bred Seed Nigeria will be marketing 8644-27 in 1992.

M.2.4 Collaborative downy mildew screening (§)

M. Fakorede (Obafemi Awolowo Univ., Ife) and S.K. Kim

Downy mildew has now spread westward to Ife, Nigeria and trials conducted by OAU scientists were heavily affected by it in 1991. In the same year, incidence of downy mildew in susceptible checks using artificial infestation in trials in Akure ranged from 10% to 70%. Considering the urgency of the DM problem and the inconsistency of present inoculation methods, evaluation of some selected lines (200) and hybrids (50) will be replicated at Ife under natural conditions in 1992.

M.2.5 Comparison of streak screening methods (§)

M. Esseh-Yovo

IITA's field infestation system has been eminently successful over the years. However it does allow about 10% escape plants under the best conditions and this can rise to 30% or more if mistakes occur. A system used in Togo using cage inoculation in the screenhouse followed by transplanting to the field, is virtually escape-proof. However it has other drawbacks (laborious, requires field irrigation, puts unrealistically severe pressure on the plant etc.) Still, it may be valuable for specific research purposes where escape-free conditions are desired. The two systems will be compared and the pluses/minuses and potential of each will be evaluated.

M.2.6 Resistance to the pink stem borer

J.G. Kling, S.K. Kim and N.A. Bosque-Pérez

Sesamia calamistis is an important second-season pest. It does most of its damage within the first several weeks of crop growth, destroying the growing point, causing stand losses. Screening of the two *Sesamia*-resistant OP pops was minimal in 1991. Work will resume in 1992. Selected S1 lines from TZBR *Sesamia*-3 cycle 0 will be recombined to form Cycle 1. Host-plant reaction to the insect appears highly dependent on plant vigor, so soil fertility microvariation (which largely determines vigor) complicates screening trials. Further, inbred lines generated in the S1 family selection process express differential degrees of inbreeding depression, and may thus perform differently because of the breeding method used (inbreeding) rather than differences in resistance *per se*, further confounding the screening process. To study this, approximately 150 S1 lines from TZBR *Sesamia*-1 C1 and their corresponding testcrosses with susceptible inbred TZi 28 will be compared under artificial infestation in the screenhouse. This will permit a cycle of selection to be completed and at the same time to determine the correlation between inbred and noninbred testing materials to separate inbreeding depression from "true" susceptibility; uninfested controls will also be used to correct for the vigor factor. Screening of inbreds/hybrids for *Sesamia* resistance will become a routine breeding activity starting 1992; TZi 4, found resistant in the early 1980's, is used as a check. Significant differences in resistance were observed under artificial infestation; 9 advanced streak resistant inbred lines were identified.

M.2.7 Resistance to the African sugarcane borer

J.G. Kling, S.K. Kim and N.A. Bosque-Pérez

Three OP populations have been developed for resistance to the second important lowland maize borer, *Eldana saccharina* which does most damage from flowering to maturity. TZBR-Eldana-1 is presently in the fourth cycle of S1 family selection. S1 families were generated in 1991 by selfing streak-resistant plants. A total of 225 S1 families from this population were artificially infested with *Eldana* (west bank, IITA-Ibadan) in the second season. Selected families will be recombined to make cycle 5, and S2's will be generated for testing in 1993. A total of 210 S1 lines of TZBR Eldana-2 C2 were similarly advanced; cycle 3 will be formed in 1992. *Eldana* screening in 1992 will include S1 lines from TZBR Eldana-3 C1. Reconfirmation of 1989 and 1990 results, which indicated progress in *Eldana* resistance, was sought by repeating the trial in 1991; data are still being analyzed. Although elite IITA inbreds and hybrids were screened in the mid-1980's in the search for resistance sources for the TZBR populations, in-depth screening of inbred lines for *Eldana* resistance was not routinely done until 1990. In that year, 6 lines and 13 hybrids showed moderate levels of resistance. In 1991, twelve lines and 24 hybrids were selected. Inbred lines including TZi 9, TZi 10, TZi 18 and TZi 35 showed moderate resistance. Most inbreds resistant to *Eldana* were susceptible to *Sesamia*, including TZi 9, 10, and 35. Resistance will be reconfirmed in 1992 and additional lines tested. New MIRT introductions from CIMMYT will also be tested in 1992.

M.2.8 Stem borer resistance on-farm (§)

H. Tijani-Eniola (Univ. of Ibadan) and S.K. Kim

Some of IITA's maize hybrids appear to be resistant to *Sesamia calamistis* and *Eldana saccharina* under artificial infestation in the screenhouse at IITA, Ibadan. The relevance of these findings to realistic farm conditions needs to be determined. Researcher-managed trials using typical farmer practices (maize-cassava intercropping, heap-sowing, hand-tillage, plus/minus fertilizer) will be conducted in the second season at Ayepe, Nigeria. Two hybrids, one thought to be resistant to *Sesamia* and the other to *Eldana*, will be compared to an adapted OP variety. Previous research at Ayepe (Mutsaers, 1991: RCMD monograph no. 4) showed that a) stand losses are a major determinant of maize yield losses in the second season, and b) about half of the stand losses are due to borers, particularly *Sesamia*. Thus, reduction in stand losses caused by *Sesamia* deadhearts will be considered as evidence of useful borer resistance. *Eldana* stalk breakage and ear damage will also be measured.

M.2.9 Breeding for weevil resistance

J.G. Kling, D.K. Kossou, S.K. Kim and N.A. Bosque-Pérez

Storage pest damage is a serious problem in the forest. Research has shown that longer, tighter husks help protect maize cobs from invasion by weevils. Many improved maize populations have poorer husk cover than traditional varieties, so selection for good husk cover is now routinely practiced in all populations, and reasonable progress is being achieved. Hard, flinty kernels are also more resistant to weevils, but some forest zone consumers prefer floury (soft endosperm) maize varieties because they are easier to mill and give higher flour yield. Thus, grain hardness is not a widely useful resistance mechanism. Methods for controlled infestation of ears with and without husks have been worked out and will continue to be applied on forest-targeted populations and hybrids in 1992.

M.2.10 Screening inbreds against *Sitophilus* weevils (§)

D.K. Kossou (Univ. of Benin) and S.K. Kim

Approximately 50 inbreds and 10 hybrids will be tested for resistance to *Sitophilus* weevils under farmer storage conditions (with husk and grain on cob, in bins) in collaboration with Benin NARS scientists.

M.2.11 Improving grain quality for Benin (§)

C.G. Yallou, D.K. Kossou (Univ. of Benin), J.G. Kling and A.E. Okoruwa

Although IITA varieties have been shown to outyield traditional varieties by a large margin, Benin farmers prefer the ease of milling and high flour yield of the traditional variety 'Gbogbe' (M. N. Versteeg). This collaborative project aims to transfer the Gbogbe grain quality into the higher-yielding, agronomically-superior background of TZSR-W-1, while simultaneously improving the latter's husk cover to better protect the grains from weevils. Full-sib families from Gbogbe x TZSR-W-1*2 (backcross 2) are being generated at Ibadan during the dry season. These will be sent to Benin for field trials in 1992 where selections will be made for husk cover and weevil resistance. Remnant seed from the best families will be increased in Ibadan in 1993 and recombined to generate the next cycle for further improvement.

M.3 Maize Improvement for the mid- to high altitudes

Project rationale

Midaltitudes are areas from 800 to 1500 meters above sea level, while high altitudes are those above 1500 m. Maize is important in the mid- to high altitudes of Zaire, Cameroon, Togo, Nigeria, Guinea-Bissau and Guinea-Conakry, as well as in Eastern and Southern Africa. Yield potential of this environment is higher than in the lowlands, because cooler temperatures extend crop duration without inhibiting photosynthesis, resulting in greater total carbohydrate accumulation over the season. Cooler temperatures also inhibit breakdown of soil organic matter, so these soils often have higher organic matter content and cation exchange capacity (nutrient retention capability). Volcanic-derived soils have better structure than lowland soils but are often P-fixing. Special pest organisms are found in the midaltitudes to which lowland germplasm is susceptible: *Exserohilum turcicum* blight, *Puccinia sorghi* rust, *Diplodia* ear and stalk rot, and *Phaeospora* and *Cercospora* leaf spots. *Busseola fusca* is a stem borer common to this zone. Constraints in common with the lowlands include N deficiency, drought, streak virus and soil acidity. Lowland germplasm grows poorly in

the mid- to high altitudes; different germplasm bases are needed for these environments, and have been created based on materials from the East/Southern African midaltitudes and subtropical materials from CIMMYT. The high yield potential of the midaltitudes opens a special opportunity to exploit hybrid vigor.

Completed studies

Everett, L.A., J. Eta-ndu, M. Ndiro, I. Tabi and N. Beninati (unpubl. research monograph in process). Maize breeding for the Highlands of Cameroon: populations, inbreds and hybrids from the IITA-NCRE-Cameroon program 1984-1991.

The USAID-funded National Cereals Research and Extension Project (NCRE) in Cameroon (jointly implemented by IITA and IRA) has focused on the maize-dependent Western Highlands since the early 1980's. The improved variety 'Kasai' was introduced by the NCRE project in the early 1980's from Zaire and remains in high demand in the Western midaltitudes for its flinty grain (resistance to storage weevils), somewhat early maturity and short, lodging-resistant plant type. 'Shaba', also introduced from Zaire, has also been well accepted in the Adamaoua Plateau. The latest open-pollinated midaltitude materials developed by the project (Syn 4) outyield the widely grown improved variety Coca by 10% under good research station management (6-10 ton yield level). The best hybrids add another 25% yield increment. These materials were tested in the additional midaltitude environments of Nigeria and Zimbabwe in 1992. Cameroon materials were susceptible to stalk and ear rots in Nigeria; Nigeria-developed materials were susceptible to rust in Cameroon. Both grew far too tall in the fertile, brighter Zimbabwe environment. They are clearly better-adapted than Zimbabwe material to the poorer soils and lower light intensity of West/Central Africa, particularly with regards to resistance to the major diseases (midaltitude blight, rust, stalk/ear rots and streak virus). The best materials will be registered in Crop Science, with seed samples stored in long term cold storage at IITA and in the USDA Colorado seed storage facility. Adapted materials with high combining ability and disease resistance will be merged into the Nigeria-based IITA midaltitude program.

Kim et al. (unpubl.). Genetics of resistance of maize inbred lines to *Exserohillum turcicum*.

Genetics of resistance to *E. turcicum* was studied in 1990-91 using diallel crosses (10x10) and generation mean analysis. Trials were conducted under artificial infestation of *E. turcicum* at UTC-Tenti, near Jos, Nigeria. High, uniform infestations were obtained in all trials and blight symptom ratings were taken two weeks after flowering. Preliminary analysis indicates that resistance is polygenic and additive.

Activities

M.3.1 Midaltitude breeding in Nigeria

S.K. Kim

Three testing sites on the Jos Plateau of Nigeria were used in 1991: Zalette-BARC farm (900 m), Vom-WAMCO farm (1,300 m) and UTC-Tenti farm (1,350 m). Among the OP's, population improvement of TZMSR will receive emphasis; 1992 will be the first year of full sib testing. Superior families will be used to form an experimental variety. Full-sib family selection continues in the TZMSR-W population. Progress in several traits over the four cycles of selection was demonstrated in 1991; the "across sites" experimental varieties have been the most consistent performers. Inbred and hybrid development continues, deriving inbreds from the TZMSR population. UTC Corporation is starting commercial inbred multiplication for seed production. Since the Cameroon NCRE project is winding down (see M.3.2), efforts are underway to integrate its germplasm into the Nigeria-based program. The best-performing materials identified in 1991 from Cameroon and holding *E. turcicum* and *Diplodia* stalk/ear rot resistance in Nigeria in 1992 will be test-crossed with inbred testers TZMi 101 and 407 to classify them into the heterotic groups of the Nigeria germplasm base. Comparative yield-testing of hybrids from both countries will continue for a second year in Nigeria in 1992.

M.3.2 Mid- to high altitude breeding in Cameroon

N. Beninati (IITA-NCRE), M. Ndioro and I. Tabi (IRA),
and J. Foko (Univ. Centre Dschang)

Maize is the dominant staple food crop in the Western Highlands of Cameroon, the most heavily populated part of the country (25% of total population), accounting for 60% of the country's maize production. The NCRE highlands breeding project has been improving maize for this zone since 1984 and will continue to 1994, although the work is winding down due to budget cuts. Five trial locations are currently utilized in the Western Highlands: Foubot (1,000 m), Babungo (1,200 m), Bambui Station (Mfonta site, 1,300 m), Dschang (University Centre Dschang, 1500 m) and Mbiyeh (MIDENO Station, 2,100 m). Maize is also a new high-

potential crop in the Central Highlands (Adamaoua Plateau); an additional site is located there, called Mbang Mbirni (1,200 m). Half-sib family selection is underway in TZEMSR-W, ATP (acid tolerant population), EWP (early white population) and HAP (high altitude population). Several populations used as germplasm sources are also being maintained or improved in the midaltitudes, e.g. Ecuador 573 and Kitale 2 from Kenya, Kasai from Zaire, Populations 90 and 92 from Tanzania, and Populations 32 and 43 from CIMMYT. Streak resistance conversion is underway in Shaba and Kasai; two screenhouses were installed in early 1992. The "comprehensive breeding system" used in Cameroon has produced both high yielding OP synthetic varieties and hybrids. The latest synthetics, Syn 4 to Syn 6 show substantial yield increases over previous material (about 10%). Inbred and hybrid development continues with selfing from populations, crosses, and reciprocal synthetics. Inbreds and hybrids are in preproduction testing by Pioneer Hi-Bred International.

M.3 Near-midaltitude maize for Togo (§)

M. Esseh-Yovo and S.K. Kim

A small area in western Togo (Dayes and Gbadi-Nkugnan sites) approaches midaltitude (700-800 m) and lowland varieties do poorly there. IITA's TZMSR and midaltitude hybrids were clearly superior in 1991 and will be tested again more rigorously in 1992.

M.4 Maize germplasm enhancement

Project rationale

Continuing progress in maize breeding will depend on enhancing the gene pool. Desirable qualities may be found in exotic germplasms, but these are usually maladapted to African environments. They need to be incorporated into adapted breeding populations.

Completed studies

Kling, J.G. and N. Q. Ng (unpubl.). Characterization of African maize landraces.

Small samples of 584 landraces were given to maize breeders in 1989, representing Kenya (49 accessions), Equatorial Guinea (7), Zimbabwe (49), Cameroon (6), Chad (84), and Togo (97). The accessions were planted in two replications and inoculated with *Bipolaris maydis*. Plant-to-plant crosses were made within rows to increase seed stocks to permit additional screening for other biotic constraints. As additional seed increases are made, samples will be returned to the Genetic Resources Unit for long-term storage.

Kim, S.K. (unpubl.). Screening of wild relatives of maize for resistance to *Striga hermonthica*.

Five *Tripsacum* spp. from Univ. of Massachusetts (Prof. W. Gualinat), 5 teosinte accessions from CIMMYT's germplasm bank, and 3 *Zea diploperennis* introductions as well as some lines derived from *Zea diploperennis* x *Zea mays* crosses from Univ. of Hawaii (J. Brewbaker) were challenged with *Striga hermonthica* in screenhouse pots in 1988-90. None of these was immune, but some showed reduced striga emergence. These results are preliminary.

Activities

M.4.1 Screening landraces against striga

J. G. Kling and D. Berner

About 500 American landraces from CIMMYT's germplasm bank and 500 African landraces held at IITA have been increased and are being evaluated under *Striga hermonthica* infestation at Mokwa. Infestations were suboptimal in 1990 and 1991; 1992 will be the third year of testing. Lines that show few or mild symptoms of striga attack and/or little or no striga emergence will be reevaluated in pots in the screenhouse for reconfirmation. Promising materials will be selfed and reselected in order to concentrate resistance genes, and will then be introgressed into adapted STR populations.

M.4.2 New sources of striga resistance (§)

M.D. Winslow, D.K. Berner, S.Y.C. Ng and Y. Savidan (ORSTOM-CIMMYT)

Recently, ORSTOM scientists posted to CIMMYT have enlarged the collection of *Tripsacum* and are studying ways to transfer genes from *Tripsacum* to *Zea mays*. This could provide the maize gene pool with access

to a larger, more diverse gene pool than previously available. Approximately 30 representative collections will be imported, germinated through embryo culture and tested for resistance against striga in greenhouse pots.

M4.3. Fitting varieties to environments

J.G. Kling, A. Akalumhe, S. Jagtap, J.M. Fajemisin and A.O. Diallo

In order to refine our understanding of G x E interactions and adaptation of elite materials to different environments, a collaborative project is underway with the agroclimatology unit of RCMD (1991-1992). Current OP varieties from IITA, CIMMYT and NARS, plus one representative variety from each of the major pools and populations for which bulk seed stocks are available in our seed store, were assembled into trials. At each site there were three trials: late-maturing (25 entries), intermediate-maturing (10 entries) and early-maturing (25 entries). Locations used included several in Nigeria, Cote d'Ivoire and Burkina Faso, in 1991. Agronomic (all sites) and physiological measurements and yield components (one site, Ikenne) are being taken to identify distinguishing characteristics of these varieties. G x E analysis will be conducted to determine the range of adaptation of each variety. Additionally, data from International Trials from 1985 to 1989 has been analyzed using a G x E software package from Australia, modified by MIP. This information will be summarized in 1992 and used to establish genetic coefficients for each variety using the IBSNAT - CERES Model and agroclimatic data from the geographic information system (GIS). A comprehensive varietal and environmental information system will then be developed which will permit mapping of all testing sites and regions of adaptation for each variety. Results will be compiled in a germplasm characterization data base and fact sheets will be prepared for each variety that can be readily distributed with seed upon request from NARS.

M.4.4 Seedling-stage drought resistance (§)

M. D. Winslow

Drought at the seedling stage causes stand losses and reduced income due to costly replanting and delayed harvest. Some traditional varieties from dry areas in the Americas have long coleoptiles allowing deep planting (30 cm) to avoid the desiccation and excessive soil temperatures that occur near the soil surface. Seed samples will be obtained and tested at different sowing depths in conventional-till plots and on ridges during the dry season, when soil temperatures are highest.

M.5 Maize grain quality

Project rationale

Little research has been done worldwide on maize grain quality, since it is mainly an animal feed in developed countries. This modest project is only two years old. The first priority has been to try to identify the major parameters associated with "grain quality" for the most common end-uses in Africa. Working hypotheses have been developed and are being tested. Once these parameters are defined in specific physicochemical terms, rapid screening techniques for them will be worked out. These will then be applied to investigate the genetic control of these parameters, and to breed varieties that are acceptable for various end-uses in a high yielding, improved maize genetic background.

Completed studies

Okoruwa, A.E. and J.G. Kling. 1991. Grain characteristics related to food uses of some maize varieties. Special Annual Report, Maize Quality and Utilization Project, IITA, Ibadan; presented at Twelfth National Maize and Legumes Workshop, Kumasi, Ghana 24-26 March 1992; and in prep. for public. In J. Food Sci.

Twenty-five OP's and 11 hybrids, increased under uniform conditions at Ibadan in 1990 were tested for a large number of physical and chemical grain characteristics and starch pasting properties in 1991. Highly significant differences among varieties were found for all traits measured. Hybrid 8338-1 was lowest in oil content and highest in starch content. Low oil content is preferred for most dry milling purposes and from a storability standpoint; maize oil is not yet an important commercial product in subSaharan Africa. Varieties appeared to fall into two general categories, either high or low oil, suggesting that one or a few genes may be involved. There was little correlation between oil content and germ size, in contrast to reports in the literature for temperate maize, so selection might not have the undesirable effect of reducing germ size; this

should be investigated further. Most African food preparations involve processing of extracted starch. Wide variation found in starch pasting properties suggest that it should be possible to identify and/or develop varieties well-suited for the many different food preparations consumed in Africa, as well as for various industrial applications.

Kim, S.K. and C. Thè (unpubl.). Green maize studies of hybrid vs. OP varieties.

Twenty varieties of maize (14 hybrid and 6 OP) were harvested as green maize at 15 and 20 days after flowering (measured individually for each plot) as well as for grain at maturity for two years at two locations each in Nigeria (Ibadan and Ikenne) and Cameroon (Nkolbisson and Nutui). Yield rankings of varieties for green maize differed from those for mature grain. Maize that must withstand stresses until full maturity must have good resistance to stalk lodging, stalk and ear rots, late infected foliar diseases such as *Puccinia polysora*, etc. but these resistances are not as important for green maize which is harvested before their effects are serious. Palatability tests showed no differences between hybrids and OP's for green maize eating quality.

Osanyintola, O.J., J.O. Akingbala, and J.H. Mareck (Ph.D. thesis in preparation.). Influence of maturity, variety, and length of storage on physical, chemical, and sensory properties of green field maize.

In spite of the popularity of green maize in the forest zone of West Africa, little is known about factors that determine 'quality' to consumers. These studies showed that sugar to starch ratio and kernel hardness were important factors determining palatability. Similar observations have been reported in the literature on sweet corn.

Activities

M.5.1 Environmental effects on maize grain quality

A. Okoruwa and J.G. Kling

Results last year indicated that not only genetic factors, but also the environment in which a maize crop is grown, affects the physical and chemical properties of the grain. An experiment is underway (1991, 1992) comparing three environments across the north-south rainfall gradient (Samaru, Mokwa, and Ikenne); physical, chemical and milling properties of grains of 8 varieties grown in large, replicated plots will be evaluated to confirm and extend our understanding of this phenomenon.

M.5.2 Rapid test for milling performance

A. Okoruwa and J.G. Kling

Different milling systems are used by traditional, small-scale mechanized, and industrial operators. Several rapid tests of kernel hardness and density are being compared with results from "real-life" milling tests to assess their representativeness as screening methods.

M.5.3 Improving traditional hand milling

B. Assa Kante (IER, Mali), A. Okoruwa and J.G. Kling

Maize is more difficult to hand-mill than millet and sorghum, inhibiting its adoption in some areas. A collaborative project with IER, Mali was initiated in 1991 to evaluate varietal differences in ease and effectiveness of hand-milling, and to develop simple modifications of the traditional maize pounding method that are less labor intensive.

M.5.4 Oil content of whole vs. dehulled maize

A. Okoruwa and J.G. Kling

Oil content will be measured before and after milling to determine if oil content of whole maize is a good indicator of oil content of dehulled maize flour, an important parameter for storability and brewing quality.

M.5.5 Inheritance of grain oil content (§)

J.G. Kling and A. Okoruwa

Results in 1991 revealed that oil content of many varieties is higher than desired for most dry milling and storage purposes. Considerable genetic variation was found, and some evidence suggested few-genic control, so selection for lower oil should be possible. Inheritance will be studied using a diallel cross of inbred parents of hybrids known to span a range of oil content values.

M.5.6 Evaluating floury conversions (§)

A.E. Okoruwa and J.G. Kling

Floury-grain conversions of some improved dent and flint varieties such as TZB-SR and TZESR-W have been carried out by single-gene backcrossing to provide high-yielding modern varieties that meet the grain quality preferences of traditional consumers. However, pasting properties in these converted varieties was found to differ considerably from the unconverted version, so it seems that single gene conversion affects more than just grain density and flour yield. This study will check these conversions in comparison to traditional floury varieties to see whether they are acceptable for pasting and related properties.

M.5.7 Tortillas as bread substitute

V.A. Obatolu (IAR&T), A.E. Okoruwa and J.G. Kling

Outflow of hard currency to import wheat for breadmaking constitutes an economic drain on West African countries. In Mexico and Central America, maize tortillas play the role of bread. A collaborative project with IAR&T is in progress to a) reproduce the Mexican tortilla using locally-available materials; b) conduct village interviews and demonstrations to assess the acceptability of the Mexican tortilla; c) modify the recipe based on village reactions to make it palatable to Nigerians; d) identify maize varieties most suitable for production of the "Nigerian tortilla"; e) determine the storability of the Nigerian tortilla; and f) conduct home-level demonstrations and adoption studies to see how to fit the Nigerian tortilla into the home environment. 1990/91 research satisfactorily completed items a) and b); in 1992 work will focus on c) and d).

M.6 NARS collaboration and training

Project rationale

A major goal of IITA is to strengthen NARS research capacity. Additionally, IITA's research effectiveness can be increased through collaboration with national scientists, extending our perspective and experience, our range of sites, and our linkage to agencies that can transfer technologies to farmers. For all these reasons, collaborative activities with NARS underpin much of the research already described—consuming about 30% of MRP's budget. Activities highlighted below are explained in more detail under the relevant agro-ecosystem project.

Activities

M.6.1 SAFGRAD Network terminal phase

J.M. Fajemisin and B. Badu-Apraku

Since the mid-1970's, the SAFGRAD (Semi-Arid Food Grains Research and Development) Network has stimulated collaboration in maize improvement research among NARS for mutual benefit and complementarity. It also provided technician training and a forum for exchange of results through workshops. Collaborative research produced valuable germplasm that is in advanced testing and release stages in several countries. MIP backstopped SAFGRAD in all these activities. Officially ended on Dec. 31, 1991, SAFGRAD Phase II has been extended at a reduced activity level to December 31, 1992. During the extension, the top priority will be to assess impact from the previous Phases. A USAID-appointed specialist consultant will assist in this study.

M.6.2 International germplasm trials

J.G. Kling, S.K. Kim and S. Adewunmi

International trials, assembled in collaboration with SAFGRAD, are distributed to all interested NARS. They include both "near-finished" and "source" breeding material. NARS entries as well as IITA entries are included. IITA collates and analyzes data returned by NARS and sends them results including cross-site analysis to illuminate stability of performance across environments. An international drought testing trial will be offered for the first time in 1992, as requested by the SAFGRAD Steering Committee.

M.6.3 Usefulness of IITA germplasm in Togo

M. Esseh-Yovo (DRA, Togo), S.K. Kim and J.G. Kling

Our 1992 visiting collaborative scientist will examine the potential of IITA germplasm for maize improvement in Togo. Transfer of striga and streak resistance into Togolese material will be initiated. Combining ability

of IITA inbreds with Togolese varieties will be tested to see if topcross hybrids can be developed for commercial use in Togo. Midaltitude materials will be scrutinized for 600-800 m areas in Togo. Farmers' reactions to IITA material will be studied on-farm in Togo. Streak resistance of traditional West African varieties will be upgraded, and IITA vs. Togolese streak virus challenge methods will be compared. See also M.1.1, M.1.6, M.2.5 and M.3.4.

M.6.4 Adoption of improved maize in Nigeria (§)

M. Adenola, M. Fakorede, J. Akinwumi, J. Iken (Maize Assoc. of Nigeria);
M.D. Winslow, F. Nweke, S.K. Kim

Improved maize varieties yield 30-100% more than traditional varieties and resist the major diseases of the region. Because of these attributes and the efforts of seed multiplication and extension agencies over the years, it is believed that they have been widely adopted. However, objective data are needed to validate this impression. A 2.5 year study modeled on the COSCA format will gather information on adoption and farmers' preferences and practices. Group interviews will be held in about 250 villages and seed returned to IITA for confirmation of varietal identity.

M.6.5 Nigerian savanna maize site (§)

M. Omidiji and J. Iken (IAR&T); S.K. Kim and M. Winslow

RRPMC will provide funds in 1992 for IAR&T, Nigeria to post a breeder to the savanna at a site to be utilized jointly with IITA.

M.6.6 Savanna Station, Cote d'Ivoire

M.D. Winslow, J.M. Fajemisin, K. Goli and A. Koffi (IDESSA) and A. Diallo (CIMMYT)

Efforts ongoing since 1987 to start-up activities at the Station will continue again in 1992. Current issues are: developing the administrative and technical agreements; planning Station development; installation of a streak resistance screenhouse and training staff; setting up striga lab and training staff; and some routine breeding trials.

M.6.7 Collaborative research

1. Breeding for high yield in the savanna and forest - S. K. Kim, J. G. Kling, C. Thè and M. N'Kishama. See M.1.1, M.2.1.
2. Breeding for striga resistance - S. K. Kim, D. K. Berner, C. Thè, P.Y.K. Sallah and M. Esseh-Yovo. See M.1.6.
3. Evaluation of usefulness of IITA germplasm for maize improvement in Togo. M. Esseh-Yovo, S. K. Kim and J. G. Kling. (§) See M.5.3 and M.1.1, M.1.6, M.2.5 and M.3.4.
4. Breeding for resistance to downy mildew - J. G. Kling, S. K. Kim, K. F. Cardwell and M. Omedeji (IAR&T). See M.2.3.
5. Screening of downy mildew resistant material - Dr. M. Fakorede, Obafemi Awolowo University and S. K. Kim. (§) See M.2.4.
6. Collaborative screening for stem borer resistance under natural infestation - C. Thè, S. Twumasi-Afriyie, N. A. Bosque-Pérez and J. G. Kling. Seed from the latest cycles of *Eldana* and *Sesamia* populations were sent to selected NARS colleagues in 1991 for evaluation and incorporation into their breeding programs. This collaboration will continue in 1992.
7. On-farm testing of stem borer resistant maize hybrids - Dr. H. Tijani-Eniola, University of Ibadan; S. K. Kim and M. D. Winslow. (§) See M.2.8.
8. Effect of husk cover on postharvest performance of improved and local maize varieties - D. K. Kossou and N. Bosque-Pérez. See M.2 (completed studies).
9. Breeding for husk cover and weevil resistance - J. G. Kling, D. K. Kossou, S. K. Kim and N. A. Bosque-Pérez. See M.2.9, M.2.10.
10. Development of a variety with good dry milling and storage properties for Benin - D. K. Kossou, C. G. Yallou, J. G. Kling and A. Okoruwa. (§) See M.2.11.
11. Traditional maize processing in Mali - B. Assa Kante, A. E. Okoruwa and J. G. Kling. See M.5.3.
12. Potential of maize tortillas in Nigeria - V. Obatolu, A. E. Okoruwa and J. G. Kling. See M.5.7.
13. Collection of local varieties in Guinea and Nigeria - N.Q. Ng, F.L. Guilavogvi, El Sanonssy Bay and M. D. Winslow. (§) See G.1.3.

M.6.8 Graduate training

1. Genetic control of striga resistance in inbred lines - V. Aditimirin (Ph.D. thesis) and S. K. Kim. See M.1.6.
2. Genetic study to determine potential for improving resistance to *Striga hermonthica* using population improvement - L. Akanvou, J. G. Kling, and D. K. Berner. (§) See M.1.8.
3. Breeding for nitrogen use efficiency - H. A. Akintoye (Ph.D. thesis) and J. G. Kling. See M.1.2.

M.6.8 Training course

J.G. Kling, S.K. Kim and M.D. Winslow

The Maize Research and Technology Transfer course, held in alternate years, will take place for 8 weeks in July-August 1992. It is targeted at maize researchers and high-level research technicians. It is expected that this will be the last time the course is held at IITA, as efforts are underway to decentralize it to NARS.

Grain Legume Improvement Program

Cowpea (*Vigna unguiculata* (L.) Walp) is an important food legume in the dry savannas of Africa and tropical Asia, Africa and South America. About 80% of the world total of 7.7 million hectares is grown in the savannas of subSaharan Africa (particularly in Nigeria and Niger) where it originated, usually in mixtures with cereals with no or few inputs. Cowpea is also widely grown in Brazil and in lowland tropical Asia.

Cowpea is especially valued in the cereal-based farming systems of the African dry savanna because protein-rich plant biomass is in short supply there. Green leaves, green pods, green peas and dry grain are all consumed by people. Cattle production is a major activity in the dry savanna, so in many cases the fodder value of cowpea is greater than that of its grain. Major constraints in the dry savanna are insect pests, low soil fertility, drought and heat, striga, and fungal, bacterial and viral diseases. Cowpea has excellent tolerance to the acid soils of the forest, but insects are an even more severe problem there, so other legumes are being considered.

During the last two decades, high grain-yielding, early-maturing, erect-habit lines have been developed at IITA which have good disease resistance and improved (but still suboptimal) insect resistance. With proper management and 2-3 sprays of insecticides, these varieties can yield about 2 tons/ha within 60-70 days. Over 45 countries have released such improved varieties from IITA. However, due to uncertain availability or acceptability of insecticides, most smallscale farmers in Africa do not spray. This has limited the adoption of improved varieties, so solving the "intractable" problems of insect resistance forms a major focus of current work.

Although soybean is not yet a major crop in Africa, it has great potential in the moist savanna to play a role like that of cowpea in the drier areas. Soybean is much more resistant to insects, enabling much higher yields without insecticide sprays. It is relatively easy to grow and fits well into existing cropping systems there, breaking the continuous cereals cycle and pest buildups (eg. striga) that occur now. It is not, however tolerant of the acid soils of the humid forest.

Two major constraints, poor seed viability in storage and inability to nodulate freely with African rhizobia, were overcome by research at IITA in the 1970's-80's. A remaining major constraint is low demand for the commodity because of non-familiarity of African consumers with ways of utilizing it to make dishes in the home. This bottleneck is being addressed through a special project (described later), and exciting increases in production and consumption are occurring in some areas in Nigeria and Zambia. Demand is also increasing for industrial purposes (oil and feed) in countries like Zimbabwe.

L.1 Cowpea improvement for the savanna

Project rationale

Local varieties are often photosensitive, spreading types, grown primarily for fodder and secondarily for grain. Average yields are low (100 to 300 kg/ha) due to numerous insects and diseases, low plant populations and competition with cereals for light, moisture and nutrients. Current breeding efforts emphasize developing lines with better resistance to disease and insect pests and better growth in poor soils, more efficient recovery and use of nutrients and moisture, shade and drought tolerance, and better competing ability with cereals when intercropped, all within the spreading plant-type background that farmers prefer to use in their local cropping systems.

Completed studies

Singh, B.B. and A.M. Emechebe. 1990. Inheritance of *Striga* resistance in cowpea genotype B 301. *Crop Sci.* 30:879-881.

The parental, F₁, F₂ and backcross populations of the cross of the resistant parent B 301 with the susceptible parent IT84S-2246-4 were screened for *Striga* resistance using pot culture. The segregation pattern indicated a monogenic dominant inheritance. The gene symbol 'Rsg' (resistant to *Striga gesnerioides*) was assigned for this trait.

Singh, B.B., A.M. Emechebe and I.D.K. Atokple. 1992. Inheritance of *Alectra* resistance in cowpea genotype B 301. (submitted to Crop Science).

Genetic studies revealed that resistance to *Alectra* in B 301 is controlled by duplicate dominant genes. These have been assigned gene symbols 'Rav₁' and Rav₂ (resistance to *Alectra vogelii*).

Atokple, I.D.K., B.B. Singh and A.M. Emechebe (unpubl.). Independent inheritance of *Striga* and *Alectra* resistance in cowpea genotype B 301.

Since B 301 is resistant to *Striga* as well as *Alectra*, genetic studies were conducted to ascertain whether the resistances are independent of each other. The parental, F₁, F₂, and backcross populations of resistant x susceptible crosses were screened under simultaneous infection by *Striga* and *Alectra*. The segregation pattern revealed independent inheritance of the two traits.

Singh, B.B. and A.M. Emechebe 1991. Breeding for resistance to *Striga* and *Alectra* in cowpea. Proc. 5th Intl. Symp. on Parasitic Weeds, Nairobi, Kenya. CIMMYT. p. 303-305.

The resistant line B 301 was crossed to the susceptible but high-yielding line IT84S-2246-4 and F₁ was backcrossed to IT84S-2246-4. BC₁ F₁ plants were infested with *Striga* and only resistant plants advanced further. From the segregating F₂, F₃, F₄, and F₅ progenies, individual plants were selected which had combined resistance to aphid, bruchid, thrips, *Striga* and *Alectra* and looked similar to IT84S-2246-4. Several F₆ progenies were bulked and tested for agronomic performance. Also, these are being used as parents in a new cycle of crosses because B 301 has small seeds and other undesirable traits.

Nelson, S. (Purdue Univ.) and B.B. Singh (unpubl.). Variability for grain quality characters and cooking time in improved cowpea breeding lines.

One hundred improved cowpea breeding lines were analysed for protein, carbohydrate, fat, ash content and cooking time. Significant genetic variability for all attributes was observed. Differences in protein content, carbohydrate content and cooking time were particularly large. The results indicate the possibility of breeding cowpea varieties with improved protein content and reduced cooking time.

Activities

L.1.1 Characterizing cowpea cropping systems

B.B. Singh, D. Florini and T. Mesfin

An on-farm study (14 selected farms) was conducted in Minjibir and Gezawa local government areas of Kano State in 1991. A 20 x 20 m block was marked in each field and detailed notes were taken on field history, land preparation, crops and varieties planted, dates of planting, planting densities, planting patterns, diseases, insects, maturity, harvesting and yields of grain and fodder. This preliminary study will permit quantitative description of different systems and their constraints which will help sharpen research focus. Preliminary observations indicated that farmers intercrop two types of cowpea varieties in alternate rows with millet and/or sorghum in the same field - one for grain and the other for fodder. Both varieties are photosensitive and spreading but the grain type is earlier in maturity and planted earlier than the fodder type. Principal constraints seem to be low population, competition with cereals, diseases, insects and late maturity.

L.1.2 Cropping systems in the dry savanna (§)

M. Mamane (INRAN), S. Blade, B.B. Singh, D. Florini and H. Bottenberg

A survey of cropping systems will be done around Niamey and Maradi in Republic of Niger which is the second largest cowpea-growing country. A few selected farms at both sites will be studied in detail and notes taken on component crops in the mixture, planting densities, spacing, planting dates, diseases, insect pests, yield of grain and fodder of each component. This should illuminate production constraints in dry savannas.

L.1.3 Collection of local varieties

B.B. Singh

A number of local varieties from Northern Nigeria are being collected and evaluated for agronomic characteristics. All are photoperiod-sensitive and spreading-habit but some are earlier-maturing than others. The early-maturing ones are normally grown for grain and the late ones for fodder. Considerable variability was found for seed type and agronomic characters. Individual plants were selected from these varieties and genetic variability for agronomic traits is being studied. Samples of collected varieties will be sent to the Genetic Resources Unit.

L.1.4 Breeding value of local varieties (§)

B.B. Singh, S. Blade and NARS scientists

Local varieties differ from region to region, and vary widely for seed type, maturity and photosensitivity. To improve local varieties or to develop improved local-type varieties we need to better understand their genetic diversity. In 1992, farmers' fields around Maroua (Cameroun), Kamboinse (Burkina Faso), Bamako (Mali), Niamey and Maradi (Niger), and Sokoto and Maiduguri (Nigeria) will be visited and seed of local varieties collected and evaluated in collaboration with national scientists. Interesting material will be used in breeding.

L.1.5 Local variety x improved line crossing

B.B. Singh, D. Florini, H.W. Rossel, T. Mesfin

The best available local varieties or local-type varieties (spreading, photosensitive, late) were crossed with improved breeding lines to incorporate resistance to the major diseases and pests into the traditional plant type. F₁ plants were raised in the screenhouse and segregating populations are grown in the field. Selected progenies are being tested and selected annually for viruses, aphid, bruchid, *Striga* and *Alectra* resistance as well as traditional plant type.

L.1.6 Farmer-participatory breeding (§)

S. Blade, B.B. Singh, D. Florini, H. Bottenberg and E.C. Odion (IAR)

Breeding varieties for high input management is satisfactorily done at experiment stations. However, screening varieties for low-input farming on experiment stations may not be relevant because soil fertility levels and management practices are so diverse among farmers. This activity will attempt to involve farmers at the penultimate stage of breeding. About 100-200 g seeds of about 15-20 advanced lines will be given to 30-40 selected farmers to be grown using their own management regime, together with their preferred local varieties. One farmer will have a maximum of 2 lines, so each line will be tested on up to four farms. The plots will be monitored by researchers for agronomic performance, and farmers will be asked to advise on the strengths/weaknesses of the new lines. This activity will be initially tried around IITA Kano Station and if successful it would be expanded to other locations.

L.1.7 Improved photosensitive varieties (§)

S. Blade, B.B. Singh, D. Florini and H. Bottenberg

A number of photosensitive cowpea lines have been developed which look like traditional varieties but have added resistance to aphid, bruchid, and thrips and a shortened time to maturity. These will be evaluated in different cropping systems at different locations (on and off-station) using traditional varieties as checks. This will hopefully elucidate some of the bases for genotype x location and genotype x cropping system interactions (if any) to guide future breeding strategy.

L.1.8 Multilocal testing of advanced lines

B.B. Singh, D.F. Florini, T. Mesfin, S. Tarawali (ILCA), A.M. Emechebe (IAR)

Advanced breeding lines are yield and performance-tested annually at Kano, Samaru, Ibadan, Gumel, Maiduguri, Niamey and Maroua. It is proposed to add Ouagadougou beginning 1992 for virus resistance and a different strain of *Striga*. Six types of trials are conducted. 1) Early-maturing (60-70 days) erect and semi-erect lines for grain (as opposed to fodder). 2) Intermediate maturing (70-80 days) erect/semi-erect lines for grain. 3) Intermediate-maturing (75-85 day) spreading-type lines for grain; these were derived from crosses of traditional local photosensitive varieties with high-yielding improved lines in an attempt to combine the plant habit of the former with the aphid, bruchid, thrips and disease resistance of the latter. They are evaluated under three management regimes: pure crop with 2-3 sprays, intercrop without sprays and pure crop without sprays. 4) Dual-purpose (fodder + grain) local x improved-cross derivatives in the 85-90 day class, to replace farmers' 100-120 day varieties which yield little grain and are subject to terminal drought risk. 5) Late-maturing (110-125 days) cowpea varieties and other selected fodder species, tested in collaboration with ILCA at Kano and Kachia without any fertilizer or insecticides, mainly for increased fodder yield but grain yield is also measured. 6) New *Striga* resistant breeding lines, tested at two locations in Kano. Some entries have complete resistance to both *Striga* and *Alectra* and some have moderate to high resistance to one or both. They are in a range of plant types, from erect early to spreading-photosensitive-late.

L.1.9 Intercropping performance- selection methods

B.B. Singh

An experiment was initiated in 1991 to determine whether selection for intercrop performance in segregating populations must be done under intercropping, or can also be done in pure cropping which is easier. Two F₂ populations were planted in pure crop (with 2 sprays) and in intercrop without sprays; individual F₂ plants were selected from each population. This will continue until the F₆ generation after which the selected lines will be evaluated for intercrop performance.

L.1.10 Screening for intercropping performance

B.B. Singh and D. Florini

All breeding lines (over 300) in advanced and preliminary trials were also planted under intercropping with millet in 2 replications without insecticides sprays and evaluated for grain and fodder yield along with local varieties as checks. Much variability was observed for both grain and fodder yield in 1991; the best lines will be tested in larger plots in 1992.

L.1.11 Pest resistance breeding of local varieties (§)

B.B. Singh, Bottenberg, D. Florini and A.M. Emechebe (IAR)

Resistance to pests will be incorporated into traditional local varieties, which are readily accepted by farmers. This will stabilise their production while attempts will be made to develop new varieties with higher yield potential. In consultation with national program scientists, the 2-3 best traditional varieties from Burkina Faso, Cameroun, Mali, Republic of Niger and Nigeria will be selected as recurrent parents in a backcrossing program involving IT90K-59 as a donor for resistance traits.

L.1.12 Screening for field resistance to insects

B.B. Singh, T. Mesfin

All the breeding lines included in advance and preliminary trials (over 300) were evaluated under pure cropping without insecticide protection. Large differences in insect damage were noted in 1991; grain yield ranged from 0 to over 600 kg/ha with high fodder yield. Selected lines will be further tested in 1992 using larger plots and several locations to reconfirm these results.

L.1.13 Screening for insect resistance

B.B. Singh and T. Mesfin

Screening for insect resistance is done using a combination of field incidence and laboratory screening. Incidence of thrips, aphids, *Maruca* and pod bugs is noted in monocrop trials without insecticide. Artificial infestation with aphids and bruchids is done in the laboratory. Many lines show combined resistance to aphid, bruchids and thrips but little or no resistance to *Maruca* pod borer and pod bugs. The available thrips resistance is also low but noticeable in the field. It has been observed that lines which possess even this low level of thrips resistance plus less susceptibility to *Maruca* and pod bugs produce some grain without insecticide spray. Efforts are therefore being made to select for this combination of "resistances".

L.1.14 Screening for disease resistance

B.B. Singh, D. Florini, H.W. Rossel, A.M. Emechebe (IAR)

Entries from the advanced and preliminary trials are screened at hotspots for different diseases (scab, *Septoria* leaf spot, bacterial blight, ashystem blight etc). Screening for virus resistance is done at Ibadan in screenhouses. Entries showing combined resistance to virus, bacterial blight and other diseases have been identified. These will be further tested and used in breeding.

L.1.15 Additional pest resistance sources (§)

B.B. Singh, D. Florini, N.Q. Ng,
A.M. Emechebe (IAR) and M. Mamane (INRAN)

Resistance is available for *Striga*, *Alectra*, scab, *Septoria* leaf spot, bacterial blight and ashy stem blight in elite breeding lines. However, there is a need to identify additional sources of resistance in case new virulences arise. A large number of healthy (virus and pathogen free) germplasm lines will be screened each year for the next 4 years to identify additional sources of resistance and study their genetic control. Screening will be done at Kano, Samaru and Niamey.

L.1.16 Screening wild cowpea for virus resistance

G. Thottappilly, S. Padulosi and N.Q. Ng

The existing wild cowpea (*V. unguiculata*) germplasm (about 400 accessions) is being tested for 5 major cowpea viruses (cowpea yellow mosaic virus, cowpea aphid-borne mosaic virus, cowpea mottle virus, southern bean mosaic virus and cucumber mosaic virus) to identify sources of resistance and to better understand host-pathogen interactions and co-evolution.

L.1.17 Inheritance of blight resistance (§)

B.B. Singh and D. Florini

Bacterial blight and ashstem blight are the two major diseases in savanna ecologies. A few sources of resistance have been identified. Genetic studies will be initiated to elucidate the nature of inheritance of resistance to these diseases which will facilitate their use in the breeding programme.

L.1.18 Genetic studies in cowpea

B.B. Singh, I.D.K. Atokple and A.M. Emechebe (IAR)

To support the ongoing breeding program, basic genetic studies on male sterility, resistance and plant type are in progress. A number of male sterile lines and other mutants are being maintained.

L.1.19 Screening against *Striga* and *Alectra*

B.B. Singh, A.M. Emechebe (IAR)

All the new breeding lines (over 300) were screened for *Striga* and *Alectra* resistance in the field and greenhouse, with known resistant lines like B 301 and IT82D-849 as checks. The objective was to identify additional sources of resistance so that a program for breeding for horizontal resistance could be initiated. A number of lines showed moderate levels of resistance and these will be tested further at several locations for reactions to different strains of *Striga*.

L.1.20 Genetics of *Striga* and *Alectra* resistance

B.B. Singh, I.D.K. Atokple and A.M. Emechebe (IAR)

Systematic genetic studies are in progress to elucidate the allelic relationship between the genes conferring resistance to *Striga* in B 301, IT82D-849, SUVITA-2 and IT81D-994. This will facilitate the pyramiding of different genes to obtain a more durable resistance.

L.1.21 Resistance to *Striga* over time

B.B. Singh, I.D.K. Atokple and K.F. Cardwell

An experiment was initiated in 1990 in which resistant and susceptible lines along with fallow and intercrop treatments are planted in the same plot year after year. *Striga* population is assessed by planting "windows" of a susceptible variety and also by counting *Striga* seeds in the soil. The experiment will be carried for 4 years to ascertain the effect of resistant varieties and other treatments in reducing the *Striga* population.

L.1.22 *Striga* yield loss assessment

B.B. Singh and I.D.K. Atokple

A set of isogenic lines have been developed which differ with respect to *Striga* resistance. The performance of these lines is being studied with different levels of *Striga* infestation to assess yield reduction due to *Striga*.

L.1.23 New biotypes of *Striga gesnerioides*?

G. Myers and B. B. Singh

In 1990 there was a report of a strain of *Striga gesnerioides* in Benin that could overcome the resistance of B 301. Observations in 1991 supported this finding but were not completely conclusive. More refined testing of B 301 and lines derived from crosses with it will be undertaken in 1992 to verify this result.

L.1.24 Evaluation for grain quality B.B. Singh, S. Nielsen (Purdue Univ.)

There is no planned programme to breed for higher protein content and other quality characters in cowpea but efforts are made to analyse advanced breeding lines for protein, fat, carbohydrate, minerals and cooking time so that lines with below average composition can be eliminated. The results have shown that most of the improved lines have more than 25% protein and relatively short cooking time.

L.1.25 Breeding cowpea for grain quality (§) B.B. Singh, S. Nielsen (Purdue Univ.) and M. Bokanga

Considerable genetic variability has been observed for protein content (22 to 32%) and cooking time (21 to 62 min.) in cowpea. Therefore, a breeding program will be initiated to develop cowpea varieties with higher protein content and reduced cooking time.

L.1.26 Cowpea performance under irrigation B.B. Singh

Several countries in the savanna region have developed irrigation facilities where wheat or vegetables are being grown in the dry season. Cowpea can be an alternative crop. Most of the farmers with irrigated land have means to provide minimum inputs required for a good cowpea crop. A trial consisting of 10 cowpea varieties was planted at 3 dates January 19, January 31, February 20, 1991 at Wudil, Kano State in collaboration with Hadejia Jamaare River Basin Authority to identify cowpea varieties suitable for planting in fields where wheat could not be planted on time. This will be repeated in 1992.

L.2 Wide crossing for insect resistance in cowpeas

Project rationale

Reduced susceptibility to post-flowering insects such as thrips, *Maruca* pod borer, and pod bugs is being utilized through conventional breeding. However the need for spraying could be eliminated if high resistance could be achieved. Since high resistance is not found within the cultivated cowpea gene pool, we are exploring wide crosses. These have not been easy, so we are looking at biotechnological possibilities for assisting the process, in collaboration with several advanced laboratories. Conventional breeding techniques such as embryo rescue, polyploidization and cytogenetics are described here; those involving molecular biological techniques are described in the Biotechnology Research Unit section.

Completed studies

Chiappini-Carpena, P., G.O. Myers and L.E.N. Jackal (unpubl.). Evaluation of resistance to *Callosobruchus maculatus* (Fab.) and *Bruchidius atrolineatus* (Plc.) (Coleoptera: Bruchidae) In pods of cowpea *Vigna unguiculata* (L.) Walp. from field infestations.

Field infestation by *C. maculatus* and *B. atrolineatus* was studied on seven cowpea genotypes in three environments. Infestations differed in intensity at the different sites and on different genotypes. *B. atrolineatus* is essentially absent during the first season at Ibadan but is important in the second season at Ibadan and Kano. Parasitization of bruchid eggs by *Uscana* spp. was found to be significant.

N. F. Agwaranze, N.Q. Ng and T.A.O. Ladelinde (unpubl.). Genetics of pod pubescence in *Vigna vexillata*. F1, F2 and backcross progenies were created for six crosses involving 5 genotypes of *V. vexillata* that exhibit variation in pod hair density and length. Generation mean analysis will be used to determine inheritance. Analysis of data is near completion.

Activities

L.2.1 Cowpea wide cross research group (§) G. Myers, S. Schnapp, T. Mesfin, L.E.N. Jackai, G. Thottappilly, N.Q. Ng, S. Padulosi, H. Mignouna, R.E. Ugborogho, M. Tamo, and external collaborators at Purdue and several Italian Universities

This multidisciplinary Research Group will be formed in 1992 to bring a comprehensive approach to solving the insect resistance problem in cowpea. Organizational meetings will be held, a coordinator chosen and *modus operandi* worked out. Conventional efforts continue at IITA on wide crossing, embryo rescue and cytogenetics. Collaborative activities are underway with our Biotechnology Research Unit and with various advanced labs, notably the University of Napoli, Italy; the Germplasm Institute of Bari, Italy; and Purdue University, USA, to hopefully overcome some of the difficulties encountered in getting successful hybrids and gene transfer.

L.2.2 Outcrossing in wild cowpea

N.Q. Ng and J. Apeji

Spatial separation of the anther from the stigma in wild cowpea appear to cause outcrossing. Further study of this character and its genetic control is ongoing.

L.2.3 Pubescent cowpeas for insect resistance

N.Q. Ng and T. Mesfin

A new variant of wild cowpea with very hairy pods, leaves and stem which may discourage insect attack has been successfully crossed with cultivated cowpea. Hybrid progenies were advanced and good plant types with and without pubescence were selected. Fresh pods of some F5 selections were tested against *Maruca* pod borers and preliminary results indicate that a few of the hybrids show better resistance than IITA's best cowpea breeding lines (although none of the progenies is as resistant as *V. vexillata*). Tests of the progenies against other insect pests are in progress.

L.2.4 Crossability of cowpea with *V. vexillata*

N.Q. Ng, J. Apeji and S.Y. Ng

Breeders are interested in *V. vexillata* because of its resistance to cowpea pod borers and pod sucking bug, major constraints for cowpea. However, previous research on crosses between cowpea and *V. vexillata* revealed abortion at the global stage of the embryo. Between October 1990 and the middle of 1991 a mass hybridization program between two genotypes of *V. vexillata* and two of cowpea (*V. unguiculata*) was attempted and embryo rescue was tried. A total of 11,771 pollinations were made and only 183 pods remained on the peduncles for more than five days. Embryos were excised from seeds of any pods that survived at least five days. Some putative hybrids were successfully cultured from the excised young embryos. Unfortunately, later studies of the mature plants from those cultures could not confirm true hybrids.

L.2.5 Interspecific hybridization in *Vigna*

S.R. Schnapp, G.O. Myers, N.Q. Ng, S. Padulosi, S.Y.C. Ng at IITA;
P.M. Hasegawa, R.E. Shade, L.L. Murdock, L.W. Kitch at Purdue Univ.;
F. Saccardo, A. Del Giudice, L. Monti at Univ. of Napoli

Wide crosses between related *Vigna* species have been made (eg. *V. oblongifolia* accession TVnu 37 x *V. luteola* accession IRFL 6192) and embryos of putative hybrids have been rescued. F1 plants have been evaluated by morphological comparison and by DNA fingerprinting using the nuclear-encoded rDNA gene from flax as a probe. Some of these hybrids are fertile and F2 plants have been obtained. Some F1 plants have been successfully backcrossed to both parents. Successful interspecific hybrids with cultivated cowpea have not yet been achieved, however. Attempts are underway to complete the bridge cross by hybridizing the progeny of the *V. oblongifolia* x *V. luteola* cross with cultivated *V. unguiculata*. In addition, efforts are continuing to hybridize *V. luteola* with cultivated *V. unguiculata* and its wild subspecies. To date, these crosses have resulted in fertilization and pod retention to about 12 days but embryo abortion apparently takes place at about 3 days after pollination. Efforts to rescue embryos using a combination of ovary culture/embryo culture techniques are ongoing.

L.2.6 Embryo rescue in *Vigna* hybrids (§)

S.R. Schnapp

Further efforts are needed to complete the bridge cross to cultivated cowpea, through a continuation of the crossing program in combination with pod culture and embryo rescue techniques. Ordinarily, crosses between progeny of the *V. oblongifolia* x *V. luteola* hybrid and *V. unguiculata* or between *V. luteola* and *V. unguiculata* may result in fertilization and pod retention for up to 12 days, but embryo abortion occurs at a stage

when the embryos are too small to be successfully excised. Therefore, methods will be developed to improve embryo development by first culturing the intact ovary until the embryos have developed sufficiently for excision. This involves finding an appropriate culture medium and preventing microbial contamination associated with the pubescence of the pods. Physiological factors affecting flowering and pod retention will also be investigated.

L.2.7 Polyploidy in cowpea and *Vigna vexillata*

N.Q. Ng, J. Apeji and A.E. Adegbite

Attempts were made to induce polyploids in cowpea (*V. unguiculata*) and *V. vexillata* in 1991 to enhance prospects for hybridization between the two species. Four day-old pregerminated seedlings and whole seeds of one genotype each of cowpea and of *V. vexillata* were soaked in colchicine at different concentrations. Shoots of about 2-3 weeks old seedlings were also treated with the chemical. Many forms of mutants were observed. Doubling of chromosome numbers was detected in a few flower buds from plants treated with colchicine. Further investigation is underway and the experiment will be repeated. If polyploids are obtained they will be used for interspecific crosses. This work will also link up with the project being planned by Dr. Ughborogho, a visiting scientist to the Biotechnology Research Unit.

L.2.8 Tetraploid induction in *Vigna*

R.E. Ughborogho, S.R. Schnapp, F. Saccardo (Univ. of Naples),
G. Thottappilly and N.Q. Ng

An alternative approach for achieving interspecific hybridization involves performing the cross at the tetraploid level. Colchicine treatment has been used to obtain putative polyploids of *V. unguiculata*, *V. oblongifolia* and *V. luteola*. Colchicine treatments from 0.1 to 0.5 % applied to axillary buds resulted in a number of phenotypically abnormal plants. Seed is being collected from plants suspected of being polyploid on the basis of stomatal size and number, and cytological examination will be performed to confirm polyploidy. Seed treatment with colchicine is also being studied on three lines of cowpea and two wild species, *V. vexillata* and *V. luteola*.

L.2.9 Hybridization of *Vigna* tetraploids (§)

R.E. Ughborogho, S.R. Schnapp, G.O. Myers, and N.Q. Ng

Crosses will be attempted between putative tetraploids (generated in previous studies) identified on the basis of increased stomate size and reduced stomate density per unit leaf area.

L.2.10 Cytogenetics of putative cowpea polyploids (§)

R.E. Ughborogho, N.Q. Ng, G. Thottappilly and F. Saccardo

Cytogenetic confirmation of chromosome numbers will be carried out on putative polyploids. It may also be possible to identify aneuploids that may be useful in genetic studies. Further, a survey of the somatic chromosome number, size and morphology of selected accessions will be undertaken to facilitate interspecific hybridization. Karyotype differences responsible for incompatibility and embryo abortion may be detected.

L.2.11 Breeding cowpea against pod-sucking bugs

G. Myers, L. Jackai, T. Mesfin

Initial selection of material resulting from crosses of elite but susceptible lines to resistance sources was done in 1990. Further selections were made in 1991 under spray/no spray conditions and additional crosses were made. Material from the first round of crosses is now in preliminary yield evaluation and will be undergoing aphid, bruchid and virus screening. Material from the second round of crossing will be in preliminary field evaluation. Additional crosses will be made in 1992.

L.2.12 Genetics of resistance to pod-sucking bugs

M. Ogbaji, G. Myers, L. Jackai, T. Mesfin and S.R. Schnapp

The hemipteran pod sucking bugs *Clavigralla tomentosicollis* (Stal) are among the most damaging post-flowering pests of cowpea, particularly *C. tomentosicollis*. Two highly susceptible and 5 resistant lines were crossed in 1991 and progeny advanced during the first half of 1992. F₃ and BC₁F₂ progeny will be field tested at Mokwa under low and high *C. tomentosicollis* pressure. Additional laboratory screening will also be done to verify field results.

L.2.13 Pyramiding insect resistance genes (§)

G. Myers, L. Jackai and B.B. Singh

Several *V. unguiculata* genotypes have been identified with reduced susceptibility to both *Maruca* and the pod sucking bug complex. While the exact mechanisms involved are not known, it is assumed that pooling them via a convergent crossing program would boost resistance. Regular sampling of the population as it is developed and of the terminal population should provide material for a study of response to selection.

L.3 Special-purpose cowpeas and other legumes

Project rationale

Cowpea has some interesting and economically-important special-purpose niches that are addressed through modest breeding activities. Erect-type cowpeas still give much higher grain yields than traditional spreading types, to those farmers able to purchase and use insecticides. Bush-type cowpeas for fresh pod consumption (vegetable cowpeas) are important as a cash crop in Asia and Central/South America. And there is interest from the RCMD in developing live-mulch cowpeas (spreading types) for soil conservation and weed control in intercropping situations. Additional food legumes need to be identified for the forest zone.

Activities

L.3.1 Breeding erect-type cowpea

G. Myers, L. Jackai, D. Florini

The longstanding effort on the development of high yielding, disease resistant, less insect-susceptible (requiring insecticide), early and medium maturity erect-type cowpeas for grain production will continue in 1992 using the pedigree method.

L.3.2 Registration of improved cowpea varieties (§)

Over 45 countries have tested, selected and released improved erect-type cowpeas that originated from IITA. These varieties are IT82E-9, IT82E-16 IT82E-32, IT82D-889, IT83S-818, IT84S-2246-4, IT85D-3577 with 60-70 days maturity and IT81D-1137, IT81D-985, IT81D-994, IT85D-3516-2 with 75-80 days maturity. Detailed characterization of these varieties will be done in 1992 to enable publication in the form of germplasm registration.

L.3.3 Breeding vegetable-type cowpea

G. Myers, L. Jackai and D. Florini

Since we retain the world mandate for cowpea, our modest activities on this will continue in 1992.

L.3.4 Live mulch potential of cowpea (§)

G. K. Aluko (Univ. of Ibadan), G. Myers, M. Akoroda and members of agro-ecosystem Research Groups

There is interest in the potential of various legumes (including cowpea) as cover crops to suppress weeds, combat erosion and contribute nitrogen for cassava production. Trials will begin in 1992 to compare cowpea with other ground cover crops under sole and intercrop conditions. If cowpeas show promise an improvement activity will be initiated for this end-use.

L.3.5 Genetic linkage analysis in cowpea

G. Myers and B. Kehinde

Numerous morphological mutants of cowpea have been identified over the years. Studies of inheritance of several of these are underway: branching peduncle, non-petiolate, septa foliate, and crinkled leaf. Parents, F₁'s, and segregating progeny in the F₂ and BC₁F₁ will be scored for these and other traits.

L.3.6 Adaptive testing of various grain legumes (§)

K.E. Dashiell, G. Meyers, N. Q. Ng and
members of the agroecosystem Research Groups

The agro-ecosystem research groups have identified a need for additional types of grain legume in certain environments where cowpea and soybean are not well-adapted. Alternative legumes to be tested include pigeon pea, groundnut, yam bean, lima bean, wing bean and bambarra groundnut.

L.4 Physiology of adaptation in cowpea

Project rationale

Most traditional cowpea varieties are slightly to highly photo- and thermal- sensitive. Presumably, these are evolutionary adaptations to wide annual variation in length of the rainy season, helping traditional varieties to minimize their exposure to heat and drought stress. Intercropping with sorghum and millet is another common feature of cowpea cultivation, and farmer selection has likely favored types best suited to this, although the specific attributes involved are not well known. A better understanding of these factors and their genetic control would enable conscious breeding efforts to utilize and improve them. Physiology work was begun in 1990 to advance our understanding in these areas. A modelling approach is being used to pull together these variables to explain yields of biomass and grain in different environments and cropping systems. Screening for drought tolerance is also underway and showing signs of progress.

Activities

L.4.1 Predicting time of flowering in cowpea

P. Craufurd and R.J. Summerfield (Univ. of Reading)

Choosing varieties with appropriate crop duration for different lengths of the growing season is essential to maximise potential production and minimise the risk of drought. Time to flowering in cowpea is determined in a quantitative manner by temperature and photoperiod, and models exist describing this. Experiments are continuing in Kano to test these models and a proposed screening technique. Approximately 25 cowpeas of varying sensitivity to photoperiod are grown in long and short daylength treatments in different mean temperatures. A set of data to test the predictions from the above experiments, at sites from Ibadan to Niamey, has been obtained.

L.4.2 Photoperiod inheritance study (§)

G. Myers and P. Craufurd

This study is to investigate the genetics of photoperiod sensitivity. It will be conducted utilizing segregating populations derived from crosses of photoperiod sensitive traditional varieties with photoinensitive, early-maturing grain types at Kano and Ibadan. Understanding the genetics of photoperiod sensitivity should allow plant breeders to more efficiently develop sensitive and non-sensitive varieties.

L.4.3 Cardinal temperatures for germination

R.H. Ellis, P. Craufurd, R.J. Summerfield
and E.H. Roberts (Univ. of Reading)

The cardinal temperatures, base (T_b), optimum (T_o) and maximum (T_m), which determine response to temperature during germination have not been adequately quantified in cowpea. Variation in these parameters, particularly T_m , may be useful in selecting for heat tolerance. Approximately 60 varieties/lines will be studied by an MSc student at Reading University.

L.4.4 Physiology of erect vs. prostrate cowpeas

P. Craufurd

Experiments have been conducted over three seasons with an erect variety IT84S-2246, and in one season with a prostrate variety 'Dan Illan', using density, sowing date and irrigation treatments to create variation in growing conditions. Variation in biomass and grain yield, yield components, radiation interception and use, water use and biomass accumulation and partitioning will be examined. Further studies will describe relations between yield and water use, and yield and nutrient use, to give a comprehensive data set for an erect and a prostrate variety useful for simulation modelling.

L.4.5 Yield and water use of cowpea

P. Craufurd

Four to six early/erect or early/prostrate varieties will be subjected to a range of water deficit treatments at Minjibir using irrigation. Treatments will concentrate on flowering/postflowering stress. Detailed measurement of shoot and root growth and water use will be made on selected treatments. In addition, a larger set of varieties will be grown with and without irrigation in Feb. - March and again with delayed sowings in Aug.-September. This will help determine if the off-season can be used for screening, given that the high temperatures and short daylengths at that time markedly alter the morphology of prostrate varieties.

L.4.6 Radiation, water and cowpea intercropping

P. Craufurd

To better understand how intercropping and environment affect cowpea growth and yield, detailed measurements of growth, radiation use and water use of sole millet and cowpea, and intercropped millet/cowpea have been made. Cowpea varieties with different phenology/morphology were grown at three locations with different total seasonal rainfall and at different density and fertility levels. Soils and plant parts will be analysed for nutrients. Crop performance ratios are being used to analyse how intercropping affects the physiology of biomass accumulation and partitioning. These experiments will be repeated in 1992 with fewer treatments, studied in more detail, and with more emphasis on below-ground factors.

L.4.7 Cowpea biomass and intercropping

E.C. Odion (IAR), R. Tabo (ICRISAT) and P. Craufurd

A number of agronomic trials in which only biomass and grain yields are measured at maturity have been carried out at Minjibir with the objective of examining how cowpeas varying in phenology and morphology perform in intercropping. Factors studied include: relative time of sowing of millet and cowpea, sowing date effects on phenology and yield in insensitive to highly sensitive varieties, intra- and inter-row spacing effects, and performance of erect and prostrate cowpeas with traditional and improved sorghums. These will be repeated for a second year in 1992.

L.4.8 Screening for shade tolerance (§)

T. Terao (TARC)

Initial studies by TARC scientists suggested that prostrate varieties were better adapted to intercropping, and that this may be due to more efficient photosynthesis at low light levels. Shading studies, combined with measurements of photosynthesis in sole and intercropped cowpea are planned to examine photosynthetic efficiency. More detailed studies on the components of the photosynthetic system will be conducted in Japan. Photosynthetic performance of a range of breeding materials will also be evaluated.

L.4.9 Screening for drought tolerance

I. Watanabe and T. Terao (TARC, Japan) and B.B. Singh

TARC scientists have screened approximately 100 lines of cowpea by establishing seedlings in the field, withholding water, and scoring for survival. Using a smaller set of these lines, a pot screening technique has been devised. The technique involves growing seedlings in 3" pots, maintaining volumetric water content at 3%, scoring for tolerance and then rewatering and scoring for recovery. A number of lines have performed well and the tests will be repeated and the best lines also evaluated in the field with irrigation. Hybridisation of the most promising material will be initiated.

L.5 Soybean Improvement

Project rationale

From 1986 through 1991 there has been a rapid expansion of soybean production in Nigeria and Zambia and other countries in Africa have initiated research and extension projects on soybean. As might be expected, the increase in soybean production has also brought about an increase in soybean disease. Increased insect problems are also expected. Soybean breeding for higher yield and resistance to these pests will continue in Nigeria and a new breeding effort will be initiated in Southern Africa. Emphasis will be placed on (i) utilizing

germplasm from strong national programs such as Brazil, Thailand, India, etc. and IITA, and (ii) further improvement of elite germplasm for resistance to pod shattering, lodging, pod bugs, frog-eye leaf spot (*Cercospora*) and red leaf blotch.

Completed studies

Dashiell, K.E. and C.N. Akem 1991. Yield losses in soybeans from frog-eye leaf spot caused by *Cercospora sojina*. Crop Protection 10: 465-468.

Three soybean cultivars were evaluated for yield losses resulting from frog-eye leaf spot in field plots at 2 locations in Nigeria with a high natural infestation of *C. sojina*. Disease severity (DS) was manipulated by fungicides to ensure a range of DS ratings to provide data on the relationship of severity to yield. Mean disease severity for unsprayed plots ranged from 0.6 to 4.5 on a 0-5 scale and plots receiving 2 sprays had lower DS values ranging from 0.5 to 2.4. There were significant differences between unsprayed and sprayed plots for grain yield and 200 seed weight. Seed weight was negatively correlated with DS.

Activities

L.5.1 Soybean improvement for the savanna

K. Dashiell, L. Jackai and C. Akem

Crosses are made using parents from the IITA breeding program, Brazil and Zimbabwe and germplasm lines. The populations are advanced through F₆ using the pedigree method. Screening methods are used for identifying lines with resistance to *Cercospora* leaf spot, bacterial pustule, pod shattering, pod sucking bugs and *Spodoptera littoralis*.

L.5.2 Multilocal evaluation of soybean

K. Dashiell, L. Jackai, C. Aken, L. Bello (University of Agric, Makurdi) and T.O. Tayo (Univ. of Agric, Abeokuta)

Trials are conducted at Zaria, Abuja, Zonkwa, Abeokuta and Makurdi. The trials are organized into three maturity groups: early (90-105 days), medium (105-120 days) and late (120-135 days). They are fertilized with 50 kg of 15.15.15 and 100 kg of 0.18.0 per hectare; no pesticides are used. Data collected includes grain yield, disease ratings, pod suck bug damage, nodulation, shattering, lodging, days to maturity, seed color, seed size and seed longevity.

L.5.3 Genetics of resistance to soybean insect pests

G. Semakula-Nankina, K. Dashiell,
I. Fawole (Univ. of Ibadan) and L.E.N. Jackai

The objectives of this study are: (i) to explore the possibility of existence of both pod and seed resistance to the southern green stink bug *N. viridula* in some soybean genotypes; (ii) to investigate the mode of inheritance of pod and/or seed resistance to *N. viridula* in (i) above and to determine any linkage relationships; (iii) to identify some leaf, shoot and pod characteristics that are associated with resistance to the southern green stink bug and to the defoliator *S. littoralis*; and (iv) to identify any transgressive segregants with better resistance to the two pests in crosses of resistant x resistant genotypes.

L.5.4 Screening against the root-knot nematode

I.C. Charles, K. Dashiell, G. Atiri and C. Akem

Objectives are to screen for resistance to root-knot nematodes and SMV and to evaluate the interaction of these two pathogens on varieties that are susceptible to both.

L.5.5 Soybean breeding in Southern Africa (\$)

K. Dashiell, C. Akem, and NARS scientists in Zambia
(E. Munsanje and F. Javaheri), Zimbabwe, Tanzania and Mozambique

The objectives are to incorporate promiscuous nodulation, improved seed longevity and resistance/tolerance to red leaf blotch (RLB) into lines adapted to the high latitudes and elevations in Southern Africa. Discussions with scientists in the region and a literature review are needed to determine the most efficient strategies to be used in this work. Zambia has already requested collaboration in the following areas: exchange of germplasm, screening for RLB resistance, RLB etiology and epidemiology research, exchange visits of scientists, information exchange and joint publications.

L.6 Nodulation in IITA soybean lines

Project rationale

One of the factors contributing to the rapid expansion of soybean production in tropical Africa is the ability of the available soybean lines to nodulate freely with indigenous soil rhizobia. However, reports from farmers of low yield in the first year of cultivation have not been uncommon, suggesting possible constraints in biological nitrogen fixation. Preliminary evidence suggests that IITA soybean lines utilize a subset of *Bradyrhizobium* spp which may be very specialised, or the IITA lines may express restricted nodulation with the *Bradyrhizobium* spp. If true, variation in the magnitude and effectiveness of such rhizobia across years and environments may be constraining yields. For farmers to benefit fully from the breeding effort of IITA, the "compatible rhizobia" (especially from the cowpea miscellary) must be sufficient in quantity and quality to meet the nitrogen requirements of the soybeans when cultivated under low input systems. The IITA soybean breeding project established collaboration with the University of Hawaii NifTAL project to i) study the limits of the promiscuous nodulation trait, identified previously at IITA, to soybean production; and ii) to develop management packages to enhance nodulation and nitrogen fixation.

Activities

L.6.1 *Bradyrhizobium* spp. field populations

R. Abaidoo, K. Dashiell and K. Mulongoy

The most probable number technique is being used to establish the population size of rhizobia compatible with IITA soybean lines. Use is made of other trap hosts such as cowpea and nonpromiscuous soybean lines to help define the taxonomic subgrouping of these rhizobia. The protocol also examines whether the selection pressures that exist in breeding sites are the same as in farmers' fields.

L.6.2 Characterisation of *Bradyrhizobium* spp.

R. Abaidoo and K. Mulongoy

Rhizobia isolated from nodules from the population studies (xxx) are being examined for their cultural, biochemical, physiological, serological and genetic relationships to *Bradyrhizobium japonicum* and *Bradyrhizobium* sp. (*Vigna*). Some aspects of this are being carried out at the University of Hawaii (NifTAL Project).

L.6.3 Rhizobia for promiscuous nodulation

R. Abaidoo, K. Dashiell and K. Mulongoy

This activity is being carried out in two phases. Individual isolates are being evaluated for their nodulation and nitrogen fixation potential in comparison with standard *Bradyrhizobium* strains. The second aspect is an *in situ* evaluation of the indigenous soil rhizobia population in comparison to recommended inoculum mixtures from *Bradyrhizobium* sp. (*Vigna*) and *Bradyrhizobium japonicum*. Their nitrogen accumulation and yield potential are being compared with chemical nitrogen fertilization treatments.

L.6.4 Nitrogen fixation in IITA soybean lines

R. Abaidoo, K. Dashiell K. Mulongoy

Fifteen to twenty lines with high nodulation potential are being evaluated in the greenhouse and at breeding sites for nitrogen fixation activity. This parameter will be tested for correlation with the extent of nodulation, nitrogen concentration in above ground plant parts and yield. Simple soil inoculations in growth pouches are aimed at developing a protocol to select for promiscuous nodulation and nitrogen fixation.

L.6.5 Environmental effects on symbiosis

R. Abaidoo, K. Dashiell and K. Mulongoy

Soybean field trials with management inputs such as *Bradyrhizobium* inoculation, optimum fertilization and indigenous population enrichment will be conducted in the major ecological zones of Nigeria (essentially where soybean trials are run). Representative farmers sites will be included in the trials.

L.7 Soybean utilization

Project rationale

Soybeans are an inexpensive protein source that can balance the diets of people too poor to afford animal protein. The first phase of this IDRC supported project (1987-1990), carried out at IITA and IAR&T in Oyo State, Nigeria, showed that soybean can augment traditional legumes and oil seeds and fortify high carbohydrate foods. In the second phase (1990-1994) the objective is to develop and encourage small-scale household processing and utilization in Nigeria and Ghana. We are collaborating with four institutions in Nigeria (IAR&T, NCRI, UNN, NAERLS) and one in Ghana.

Completed studies

Osho, S.M., H. O. Ogunlape and V. A. Obatolu. 1990. Soybeans in Nigerian foods. (A booklet of 72 recipes). IITA, Ibadan, Nigeria.

Nigerian foods were formulated from whole soybeans, dehulled soybeans, raw soyflour, processed soyflour, soybean paste, soybean grits, soybean milk, extruded soyflour, defatted soyflour and soybean milk residue.

Ogunlape, H.O. and S. M. Osho. 1990. Development and introduction of improved soybean utilization technology for use in households and small scale processing enterprises in rural Nigeria. Final report of the IDRC/IITA/IAR&T Soybean Utilization Project Phase I.

This project had five main objectives: (a) document the status of soybean utilization in Oyo State, Nigeria; (b) develop household technology for preparing soy-based foods; (c) develop small-scale processing technology for soy-based foods; (d) introduce and assess the impact of household food technologies in rural areas and (e) disseminate the results of the study. A baseline survey was conducted in 1987 to document the status of soybean production and utilization in three main villages: Ikoyi, Igangan, and Ijaiye. Soybean cultivation was found to be relatively unimportant in the farming systems of farmers at the time of the baseline survey. About 17% of the farmers cultivated soybean in Ijaiye and 40% in Ikoyi, while no farmer cultivated soybeans in Igangan. According to the farmers, factors that would motivate them to grow more soybeans are a ready market (26%), multipurpose use (26%), profitability (24%), nutritional value (20%) and knowledge of its cultivation (4%). Processing effectively eliminated trypsin inhibitor activity (TIA) and reduced phytic acid and tannin to acceptable levels. Soy-fortified dishes generally had higher protein and mineral content than the non-fortified versions. Product development research fortified traditional Nigerian foods with soybean to produce soy-egusi soup, soyvita (soy + maize), soygari and soylafun (soy + cassava), soymilk, soybread, sorghum/soy biscuit, soyamusa (a plantain based baby food) etc. Studies found that dry extrusion and screwpress technologies can be used to produce inexpensive and high quality soyoil, snacks, baby foods, and breakfast foods having a high caloric density, and protein quality at least similar to the conventional brands, with a good shelf life when packaged in simple materials, and much cheaper than the same products processed by conventional methods.

Activities

L.7.1 Consumer acceptability of soy cheeses

S. Osho

Soybean cheese can be processed from soymilk using a coagulant "*Calotropus procera*". The cheese is similar to "warankasi", the local cheese. Sensory evaluation panels will determine consumer acceptability of the product prepared using different methods. Shelf life of the soybean cheese and the local cheese will also be evaluated. If the soycheese has a shorter shelf life research will be initiated to extend the shelf life.

L.7.2 Soyfufu

S. Osho and M. Bokanga

One of the most popular cassava foods in Nigeria is fufu. Various methods of incorporating soybean into fufu will be tried. The parameters that will be evaluated include chemical analysis, shelf life studies, and sensory evaluation of soy fufu.

L.7.3 Soy chocolate bar

S. Osho (IITA), G. L. Arueya and
A. A. Aina (Cocoa Industries Ltd, Ibadan)

In collaboration with Cocoa Industries Limited we are testing the incorporation of various soybean flours (extruded, raw, defatted, etc) and roasted soybean into chocolate bars. Although this may not have much impact on the improvement of nutrition, when children and adults start to think of soybean as a candy they may become more inclined to use it in other foods.

L.7.4 Soymilk for soy ice-cream and soy yoghurt

S. Osho

The raw materials needed for the production of ice-cream and yoghurt are imported. The result has been a rapid increase in the cost of ice-cream and yoghurt. Research to produce lower-cost soy ice cream and yoghurt will be done in collaboration with a company presently producing these foods using dairy products.

L.7.5 Soycoffee

S. Osho

The cost of coffee has increased drastically in Nigeria. Research will be conducted to produce an instant drink made with soybean that will simulate the taste of coffee and be acceptable to Nigerians.

L.7.6 Maize/soy tortilla (§)

S. Osho and J. Kling

Maize tortillas serve the function of bread in Mexico and Central America, but they are low in protein. The potential of a soybean-fortified tortilla will be investigated.

L.7.7 Malted maize/sorghum/soy breakfast food (§)

S. Osho and J. Kling

Malted maize/sorghum will be extruded with soybean to produce a breakfast food. Data to be collected include chemical analysis, shelf life and sorption isotherms. Acceptability and sensory evaluation will also be conducted.

L.8 NARS collaboration and training

Project rationale

A major goal of IITA is to strengthen NARS research capacity. Additionally, IITA's research effectiveness can be increased through collaboration with national scientists, extending our perspective and experience, our range of sites, and our linkage to agencies that can transfer technologies to farmers. For all these reasons, collaborative activities with NARS underpin much of the research already described- consuming about 30% of GLIP's budget. Activities highlighted below are explained in more detail under the relevant agro-ecosystem project.

Activities

L.8.1 Cowpea international trials

G. Myers and B.B. Singh

International trials are one of the primary mechanisms by which improved cowpea germplasm developed at IITA is distributed to NARS. We will continue the program of solicitation of cooperators, packing and distribution of materials, and data analysis.

L.8.2 National Cowpea Research Project (Nigeria)

G. Myers, B.B. Singh and O.O. Olufajo (IAR, Zaria)

The Nigerian Nationally Coordinated Research Project (cowpeas) multilocally tests the best lines for potential varietal release. We participate and provide technical support.

L.8.3 International soybean trials

K. Dashiell

Trials are distributed to all national programs (except Nigeria) that request them. They include 13 of our best breeding lines to be tested with two local entries supplied by the NARS. The cooperators return a copy of their data to us for analysis. These trials involve about 30 countries each year.

L.8.4 National Soybean Variety Trials (Nigeria)

K. Dashiell, P. Oyekan (IAR&T), S.O. Olafare (NCRI),

A. Uwala (NCRI), and O.O. Olufajo (IAR, Zaria)

These trials are organized by the Nationally Coordinated Research Project on soybean and are conducted at about 20 locations in Nigeria by several different universities and research institutes. Trials are packaged and results are analyzed at IITA. About 25 of our best breeding lines are evaluated in these trials each year.

L.8.5 Collaborative research

1. Cropping systems in the dry savanna. M. Mamane (INRAN), S. Blade, B. B. Singh, D. Florini and H. Bottenberg. (§) See L.1.2.
2. Breeding value of local varieties. B. B. Singh, S. Blade and NARS scientists. (§) See L.1.4.
3. Farmer-participatory breeding. S. Blade, B. B. Singh, D. Florini, H. Bottenberg and E. C. Odion (IAR, Zaria). (§) See L.1.6.
4. Multilocal testing of advanced lines - B. B. Singh, D. F. Florini, T. Mesfin, S. Tarawali (ILCA), and A. M. Emechebe (IAR, Zaria). See L.1.8.
5. Pest resistance breeding of local varieties. B. B. Singh, H. Bottenberg, D. Florini and A. M. Emechebe (IAR, Zaria). (§) See L.1.11.
6. Screening for disease resistance - B. B. Singh, D. F. Florini, H.W. Rossel, and A. M. Emechebe (IAR, Zaria). See L.1.14.
7. Additional pest resistance sources. B. B. Singh, D. Florini, N.Q. Ng, A. M. Emechebe (IAR, Zaria) and M. Mamane (INRAN). (§) See L.1.15.
8. Genetic studies in cowpea - B. B. Singh, I. D. K. Atokple and A. M. Emechebe (IAR, Zaria). See L.1.18.
9. Screening against *Striga* and *Alectra* - B. B. Singh and A. M. Emechebe (IAR, Zaria). See L.1.19.
10. Genetics of *Striga* and *Alectra* resistance - B. B. Singh, I. D. K. Atokple and A. M. Emechebe (IAR, Zaria). See L.1.20.
11. Resistance to *Striga* over time - B. B. Singh, I. D. K. Atokple (IAR, Zaria) and K. F. Cardwell. See L.1.21.
12. *Striga* yield loss assessment. B. B. Singh and I. D. K. Atokple (IAR, Zaria). See L.1.22.
13. Live mulch potential of cowpea. G. K. Aluko (Univ. of Ibadan), G. Myers, M. Akoroda and members of agro-ecosystem Research Groups. (§) See L.3.4.
14. Cowpea biomass and intercropping - E. C. Odion (IAR, Zaria), R. Tabo (ICRISAT) and P. Craufurd. See L.4.7.
15. Multilocal evaluation of soybean - K. Dashiell, L. Jackai, C. Akem, L. Bello (Univ. of Agric., Makurdi) and T. O. Tayo (Univ. of Agric., Abeokuta). See L.5.2.
16. Soybean breeding in Southern Africa. K. Dashiell, C. Akem, and NARS scientists in Zimbabwe, Zambia, Tanzania and Mozambique. See L.5.5.
17. Development of a soychocolate bar - S. Osho, G. L. Anweya and A. A. Aina (Cocoa Industries Ltd., Ibadan). See L.7.3.

L.8.6 Graduate training

1. Genetics of resistance to pod-sucking bugs - M. Ogbaji, G. Meyers, L. Jackai, T. Mesfin, and S.R. Schnapp. See L.2.12.
2. Live mulch potential of cowpea - G. K. Aluko (Univ. of Ibadan), G. Meyers, M. Akoroda and members of agroecosystem research groups. (§) See L.3.4.2.
3. Genetics of resistance to soybean insect pests - G. Semakula-Nakinga, K. Dashiell, I. Fawole (Univ. of Ibadan) and L. E. N. Jackai. See L.5.3.
4. Screening against root-knot nematode - I. C. Charles, K. Dashiell, G. Atiri and C. Akem. See L.5.4.

L.9 Ghana Grains Development Project

Project rationale

This is a bilateral special project (CIDA, Canada + CRI, Ghana) to strengthen national capability for food legume research and extension. Currently in its final Phase (III), human resource development, with particular emphasis on research management skills, receives top priority to prepare the research system to sustain itself beyond the end of the Project. In terms of crops, cowpea, soybean and groundnut improvement are addressed, in that order of emphasis. Although the Project has recommended 4 improved cowpea varieties for release, these still lack high resistance to major insect pests, so this constraint receives top attention currently. Soybean hectareage is limited at present, but it is one of the top priority crops in the government's Agricultural Development Plan. Constraints are: lack of appropriate planting methods, seed longevity in storage, threshing, seed processing and marketing. Groundnut is the most popular food legume and oil crop in Ghana but has been neglected by research in the past. Project efforts have likewise been modest, centered around identifying production constraints and suitable areas for expansion of cultivation. Introduced groundnut germplasm from ICRISAT has shown the potential to double yields.

Completed studies

Hossain, M.A., G. Atuahene-Amankwah and R. Arias. 1991. Plant establishment and yield stability of some exotic cowpea varieties in Ghana. American Society of Agronomy. Agronomy Abstracts, 1991 Annual Meetings.

In 1990, five early maturing (60-70 days) and five medium maturing (71-80 days) high yielding cowpea varieties introduced from IITA were evaluated in farmers' fields across the major agro-ecologies of Ghana (24 and 33 on-farm sites, respectively). Regression coefficients and deviation mean squares were determined for each of the varieties in order to identify stable variety(s) that perform above the average across some environments or all environments. The results of the analyses showed that only Asontem (IT32E-32) among the early maturing group and Soronko (TVx 2427) among the medium maturing group were stable and gave conspicuous yield increases averaging 11-22% over the farmers' varieties across environments.

Atuahene-Amankwa, G. and M.A. Hossain. 1991. Evaluation of some selected cowpea varieties for grain and fodder potential. Proc. Joint SAFGRAD Research Networks Workshop, March 7-14, 1991, Nlamey.

Identification of some cowpea diseases in the forest and transition ecologies of Ghana. J. K. Twumasi and M. A. Hossain. 1991. Proc. Joint SAFGRAD Research Networks Workshop, March 7-14, 1991, Nlamey.

Effect of time of harvest on grain yield of soybean in humid zone of Ghana. 1991. M. A. Hossain, P. Osei-Bonsu and B. Asafo-Adjel. Proc. 11th Maize and Legume Workshop of GGDP held on March 25-27 at Kumasi.

Activities

L.9.1 Human resource development

M.A. Hossain and CRI staff

Graduate training, technical training and visiting scientistships are used to upgrade the skills of CRI staff. Skills in research management, on-farm research, research station management, breeders seed production and storage, and communication and extension are emphasized. Training of outposted staff receives top priority in Phase III. In previous phases, these staff could not be sufficiently trained due to on-post work requirements.

L.9.2 Germplasm introduction & evaluation

M.A. Hossain and CRI staff

International trials and germplasm for cowpea and soybean from IITA are evaluated in the savanna, transition, rainforest and coastal savanna ecologies. Also groundnut germplasm from ICRISAT is evaluated. Promising germplasm is entered into multilocational testing and/or is used as parental material in crosses.

L.9.3 Varietal development

M.A. Hossain and CRI staff

13 F₂ and 2 BC F₂ populations derived from crosses made in Ghana will be evaluated for yield, maturity, resistance to major insect pests and diseases and acceptable seed characteristics. The variety development program for soybean will evaluate 15 F₆ lines for their adaptation to second season planting in southern Ghana. These will be screened for early maturity, promiscuous nodulation, enhanced seed longevity, and resistance to shattering, diseases and insects.

L.9.4 Multilocation testing

M.A. Hossain and CRI staff

Multilocal testing of soybean, cowpea and groundnut lines is conducted in the major agroecologies in Ghana. Varieties that perform consistently better than the already released varieties are then sent for on-farm testing.

L.9.5 On-farm testing of promising lines

M.A. Hossain and CRI staff

Six early and six medium maturing cowpea lines are being tested on-farm. Three promising soybean lines are being tested on farm in comparison with a released variety. These trials are being conducted only in the southern sector of the country.

L.9.6 Screening cowpea for intercropping

M.A. Hossain and CRI staff

Most farmers (at least 80%) cultivate their food crops in mixed or relay systems. Previous studies conducted by the project have shown that medium and early maturing, semi-erect cowpea varieties could perform better under intercrop situations compared to the crawling plant type which is traditionally grown in the extreme dry savannah ecologies of Ghana. A trial is being conducted to evaluate the production potential of twenty promising cowpea lines of diverse plant type and maturity in several intercropping situations: three different growth habits of cassava (low branching, mid-height branching and no branching) and three different maturities of maize (early, medium, late).

L.9.7 Screening cowpea for eating quality

M.A. Hossain and CRI staff

All lines that are identified as promising in multilocal testing will undergo palatability tests, and will be analyzed for protein content and cooking quality before they are recommended for release.

L.9.8 Storage methods for cowpea and soybean seeds

M.A. Hossain and CRI staff

The purpose of the experiment is to identify the best packaging material for maintaining seed quality in cowpea and soybean. Seeds will be saved from harvests at the Ejura (transition zone), Fumesua (forest zone), and Nyankpala (guinea savannah zone) sites. Seeds will be dried to 10-11% moisture and stored at the same location where they are produced. The storage materials that will be tested are jute sacks, cotton sacks, paper bags and polythene (0.4 ml) bags. The materials used for storing soybean seeds are cotton sacks and polythene bags. Germination tests will be conducted at 2, 4 and 6 months after harvest.

L.10 SADCC-IITA Cowpea Project

Project rationale

Begun in 1989, this Project aims to strengthen national programs in the Southern African Development Coordination Conference (SADCC) region for their capacity to carry out cowpea research. A pathologist, breeder & agronomist carry out IITA's responsibilities by collaborating closely with national colleagues. A full staff complement was not achieved until January 1, 1992. Cowpea is almost entirely grown as an intercrop with maize or sorghum in marginal agroecological zones. Principal constraints seem to be: drought stress, poor soils, insect pests and diseases, *Alectra* and *Striga*. Viruses are even more important than in West Africa.

Some smallholder needs in East Africa are different from IITA's experience in West Africa. There is demand for dual-purpose cowpeas that yield fresh leaves and green pods as vegetables, as well as grain. Cowpea fodder is also used as an animal feed.

Activities

L.10.1 Germplasm introduction & evaluation

R.A. Amable, D.M. Naik and A.L. Doto

Local and introduced germplasm (including elite lines) is being evaluated for adaptation to agro-ecologies and smallholder farming systems characterized by low inputs and intercropping. Agronomic traits, insect and disease resistance are being evaluated. Included are the IET-1 for evaluation of plant type, IET-2 for evaluation of aphid resistance, and advanced germplasm trials. The ability of cowpea to nodulate with native rhizobia is also being assessed in the IET-1 and IET-2 germplasm introduction trials using a visual rating.

L.10.2 Varietal development

A.L. Doto, R.A. Amable and D.M. Naik

Segregating material (F4-F5) from crosses received from the IITA-Nigeria program will be screened and advanced. Aphid resistance is a high priority. Yield trials of advanced lines (PYT-4) will continue. Visits to national breeders will be made to strengthen collaboration.

L.10.3 Adaptation to cropping systems

R.A. Amable, A.L. Doto and D.M. Naik

Trials are being conducted to characterize the reaction of elite germplasm to typical cropping systems and end-uses. PYT-1 looks at drought reaction, PYT-2 at dual-purpose (leaf and grain) yield, and PYT-3 at adaptation to intercropping. Another trial examines response of plant type to intercropping, to determine selection procedures for intercropping. The productivity of different intercropping systems is being compared in another set of trials (adapted from trainee-designed demo plots in a 1990-91 course) under rainfed conditions with minimal insecticide input.

L.10.4 Collaborative research

1. Collaborative testing of cowpea advanced lines in cowpea international trials. G. Myers, IITA SADCC cowpea scientist, cowpea scientists in NARS of SADCC countries.
2. Collaborative cowpea screening in Botswana, Zambia and Zimbabwe. (§). IITA SADCC scientists, with NARS cowpea scientists in Botswana, Zambia and Zimbabwe. Breeding lines and germplasm of cowpea will be evaluated that have spreading to indeterminate growth habit, with acceptable leaf quality for use as a vegetable, and improved with respect to resistance/tolerance to aphids, bruchids, bacterial blight, ash-stem blight and virus diseases.
3. Formulating protocols for cowpea evaluation in rice based cropping systems and for dual purpose (fodder-grain) production in Mozambique. (§). IITA SADCC scientist and cowpea scientist from Mozambique.

L.10.5 Graduate training

1. Influence of low soil and air temperature on yield and components of cowpea. K. Mogotsi, R. A. Amable and A. I. Robertson (Univ. of Zimbabwe).

L.11 NCRE, Cameroon Cowpea Breeding Project

Project rationale

During the early stages of cowpea research in Cameroon, emphasis was on pest management. Yield losses have been established, effective storage methods recommended, and resistant varieties to storage insect pests are being developed. The main approach of the Grain Legume Unit of the NCRE project is to develop a broad base system of research on cowpea and minor activities on other legumes. In addition to breeding for storage insect pests resistance emphasis will be put on alleviating other constraints to cowpea production: diseases (virus); striga; dual purpose (grain + fodder); seed quality (color and size); and resistance to field insect pests.

Activities

L.11.1 Germplasm evaluation

J. Detongnon, Ch. Endondo and O. Boukar

The objective is to maintain a working collection from which materials will be selected for hybridization or for varietal trials. By testing new introductions along with the best performing lines of the previous test, about 100 lines will be selected. The present collection has about 100 lines. About 20 of these were collected or introduced in 1991. This year all 100 will be evaluated for agronomic traits and reaction to pests in comparison with released varieties.

L.11.2 Varietal development

J. Detongnon and O. Boukar

The best improved lines will be crossed with selected locally adapted lines in order to improve the local varieties. F₂ populations of crosses made in 1991 will be advanced to F₃ generation. Characteristics such as yield potential, disease resistance (virus), seed quality and dual purpose (grain and fodder) will be taken into consideration for selection.

L.11.3 Varietal testing of cowpea and soybean

J. Detongnon, Ch. Endondo and O. Boukar

Several varieties of cowpea will be tested on station (Guiring) and on multilocation sites (Samgueré, and Guetale) in series of varietal trials such as early maturity, medium maturity, etc. Varieties of soybean will also be available in the International soybean nursery.

L.11.4 Cowpea line/insecticide evaluation

J. Detongnon and Ch. Endondo

The objective is to identify good performing (high grain yield and virus free) genotypes under low insecticide application, by testing lines at two insecticides regimes (no insecticide treatment and two treatments). The best 20 lines out of the 46 lines tested in 1991 will be tested in 1992.

L.11.5 *Striga* and virus resistance screening

J. Detongnon, Ch. Endondo and O. Boukar

Field testing of selected lines on *Striga* sick plots will be performed in order to identify sources of resistance under natural infestation. Observations will also be made on virus symptoms.

L.11.6 Pure-line selection for virus resistance

J. Detongnon

Single plant selections were made in 1991 in seed multiplication plots of six varieties taking into consideration yield potential and the degree of virus infestation. The selected plants will be evaluated plant to row in 1992. The best progeny rows (least virus) will be evaluated in replicated trials in 1993.

Plantain and Banana Improvement Program

Bananas and plantains (*Musa* spp.) are important food crops and sources of revenue for smallholder farmers in Africa. About 70 million Africans obtain more than 10% of dietary calories from banana/plantain, and the gross value of annual production ranks first among food crops. Banana/plantain are mainly produced by smallholders in compound or home gardens.

The cultivated types differ according to their ecoregional distribution. The plantains (*Musa* spp, AAB group) are predominant in the humid lowlands of West and Central Africa, while cooking and beer bananas (*Musa* spp, AAA group) are prevalent in the highlands of East Africa. Declining soil fertility in permanent plantations appears to encourage several of the major biotic constraints of plantain and banana: diseases (black sigatoka, fusarium wilt), insect pests (banana weevil) and nematodes.

Research on plantain at IITA began in 1973. Early work focused on agronomy, taxonomy and selection, and was carried out at Ibadan supported by a special project. In 1979, the center of plantain research was transferred to Onne Station, in the more suitable ecological niche of the humid forest zone. Investigations began on agronomy, physiology and taxonomy. Tissue culture research for *Musa* rapid multiplication and germplasm enhancement started in 1983. IITA added plantain and banana as mandate crops in 1987 and a Plantain and Banana Improvement Program (PBIP) was created in 1991 to intensify efforts on these crops. PBIP has established collaborative linkages with the International Network for the Improvement of Banana and Plantain (INIBAP) for germplasm exchange, international testing of breeding materials and training.

P.1 Developing *Musa* breeding capability

Project rationale

Musa breeding was a part-time activity until 1991. Even so, highly significant gains were achieved which opened up many possibilities for breeding across ploidy, species and subspecies of *Musa*. Based on those exciting results, IITA decided to intensify its work by hiring a full-time breeder/geneticist in 1992. There is a need at this stage to consider breeding strategies and determine the most useful methodologies for improving this little-researched crop for African conditions and constraints.

Completed studies

Vuytsteke, D., R. Ortiz and R. Swennen (unpubl.). Second division restitution in tetraploid plantain/banana hybrids.

In 4x progeny obtained from 3x x 2x crosses in which the 2x parent is the homozygous clone 'Calcutta 4', variation in black sigatoka reaction, phenotype, and growth and yield parameters has been observed. This suggests the occurrence of segregation during the modified megasporogenesis leading to the formation of 2n (=3x) eggs. This is very important with respect to the mode of production of 2n eggs and its genetic consequences. First division restitution (FDR) mechanisms for 2n egg formation are normally due to asynapsis (no pairing during meiosis) or desynapsis (falling apart of chromosomes during diplotene or diakinesis). Synaptic variants result in the non-occurrence of crossing over and therefore homogeneous 2n (=3x) gametes are expected. The fact that segregation occurs would indicate that the mode of 2n egg formation is genetically equivalent to a second division restitution mechanism (SDR). Normally SDR 2n eggs are the result of normal first meiotic division (segregation occurs), but complete omission of second division. If this interpretation is correct, the earlier assumption that plantain improvement must necessarily be limited to "diploid breeding" may be incorrect, because the formation of 2n eggs by an SDR mechanism in the 3x female parent provides a means for the occurrence of segregation and recombination in the 3x plantain genome.

Swennen, R. and D. Vuytsteke. 1991. Bananas in Africa: diversity, uses and prospects for improvement. In N.Q. Ng, P. Perrino, F. Attere and H. Zedan (eds.), Crop Genetic Resources of Africa, Proceedings of an International Conference held in Ibadan, Nigeria, 17-20 Oct 1988, IITA/IBPGR/UNEP/CNR, pp. 151-159 (1991).

African *Musa* that produce edible fruit can be grouped into three categories: plantains (*Musa* AAB group), which are predominant in the West and Central African lowland humid forest ecologies; highland beer and cooking bananas (*Musa* AAA group), which predominate in the East African mid-altitude ecologies; and dessert bananas (*Musa* AAA and AAB groups), which are cultivated throughout tropical Africa. The black

sigatoka leaf spot disease is the overriding constraint to plantain and banana production on the continent. The introduction of resistant germplasm into village homesteads, where the bulk of the crop is grown, is considered the most appropriate control strategy.

Activities

P.1.1 *Musa* germplasm working collection

D. Vuylsteke, R. Ortiz and R. Swennen

The main goal of this activity is to collect, conserve and characterize *Musa* species and cultivars with (a) black sigatoka resistance (BSR), (b) large, pendulous bunches, (c) dwarf height, and/or (d) nematode and weevil resistance, and evaluate them in the African humid forest environment represented at Onne. Conserved germplasm (> 400 accessions) includes diploid species and cultivars (*M. acuminata* -AA and *M. balbisiana* -BB), dessert bananas (AAA, AAB), plantains (AAB), and cooking and beer bananas (AAA, ABB). Over 30 sources (2x, 3x, 4x) of BSR are available in the collection. All accessions are kept in field nurseries as well as in *in vitro* tissue culture (see xxx). Characterization of the germplasm has been partially achieved. A database file including the most important descriptors is currently being developed. Duplicate *in vitro* samples and documentation are being kept at the Genetic Resources Unit.

P.1.2 *Musa* germplasm characterization (§)

R. Ortiz, D. Vuylsteke and R. Swennen

The objective of this research is to use a descriptor list to evaluate *Musa* germplasm (2x, 3x, 4x). Then, multivariate analyses (principal components and cluster) will be used for statistical grouping of the *Musa* collection and hybrids. The development of a discriminant function can also be useful for grouping hybrids. The documentation of the collection and of the hybrids will be carried out in collaboration with the Genetic Resources Unit of IITA and with INIBAP and included in a database for worldwide distribution.

P.1.3 Screening for female fertility in *Musa*

D. Vuylsteke, R. Ortiz and R. Swennen

Musa is generally considered as an intractable crop in terms of genetic improvement. However, the identification of 37 different, seed-fertile plantain cultivars (29 French and 8 False Horn) at IITA has challenged this viewpoint. High seed production rates of up to 200 seeds per bunch can be obtained when crossing plantains with BSR wild *Musa*. The average seed set ranged from less than 1 to over 20 seeds/bunch, depending on the plantain cultivar. Also, several AAA highland bananas from East Africa have produced seed upon pollination, suggesting that this important group of staple bananas could be hybridized as well.

P.1.4 Environment and seed set in plantain (§)

R. Ortiz and D. Vuylsteke

Botanical seed production in plantains and bananas is exceptionally high at Onne station and this appears to be location-specific. High seed production rates are critical to IITA's success in plantain/banana breeding. G x E interaction will be evaluated by comparing seed production rates at Onne and Mbalmayo (Cameroon) Stations. The plantain cvs. 'Obino l'Ewai' and 'Bobby Tannap', and 'Calcutta 4' will be the female and male parents, respectively.

P.1.5 Flow of breeding materials (§)

R. Ortiz and D. Vuylsteke

The proposed breeding procedures are described in Figures 1-2. Steps are discussed in the appropriate sections. These schemes form a basis for initial work, and they will be widely discussed throughout the Institute during 1992. Based on these discussions, by the end of the year it is expected that a detailed strategy will be finalized.

P.1.6 Yield trial designs for *Musa* (§)

R. Ortiz and D. Vuylsteke

The method of maximum curvature using variance and coefficients of variation from uniform yield trials will be used to determine the size of experimental units and the adequate number of replications for yield trials in plantain and banana. The plot sizes to be tested will be 1, 2, 4, 5, 8, 10, 16, 20, 40 and 80 plants. Similarly, different numbers of replications will be tested after determining the optimum plot size. The clone 'Km 5' will be used as a standard for banana; the clones included in the experiment for perennial yield potential determination (see P.5.1) will be used as standards for plantain.

P.1.7 Diploid *Musa* breeding

R. Ortiz and R. Swennen

Plantain and banana breeding has long been conducted at the diploid level due to the perceived lack of reproductive fertility at the triploid, cultivated level. An important step in conventional breeding programs, then is the production of improved diploid material to be used as male parents in both $3x \times 2x$ crosses (to produce primary $4x$ hybrids) and $4x \times 2x$ crosses (to produce secondary $3x$ hybrids). The selection of diploid material is based on both female and male fertility, BSR and good bunch characteristics. Improved diploids are multiplied for their use on a large scale in the crossing block.

P.1.8 Outcrossing rates in $2x$ *Musa* (§)

R. Ortiz and D. Vuylsteke

'Calcutta 4', a homozygous, wild diploid clone will be used to determine the outcrossing rate in $2x$ germplasm. However, this work requires the identification of traits under monogenic control and at the recessive state in 'Calcutta 4' and at the dominant homozygous state in another $2x$ clone. The field layout will consist of interplanting both clones in an isolated field. Seed will be harvested from 'Calcutta 4' and its progeny analyzed for the segregation of the marker to determine the rate of outcrossing. Alternatively, recently developed molecular markers (see P.3.6) could be used instead of conventional markers, which are few in *Musa*.

P.1.9 Male fertility in $2x$ *Musa* germplasm (§)

R. Ortiz, D. Vuylsteke and R. Swennen

The objectives of this activity are to a) determine pollen viability through microscopic studies, and b) identify the period of maximum pollen viability. Pollen stainability, using acetocarmine glycerol jelly (acgj), will be used for a preliminary screening of the $2x$ collection. Controlled crosses ($2x \times 2x$ and $3x \times 2x$) will be performed using pollen samples collected during different stages of male flower development. Pollen samples will be stained with acgj before their utilization in crosses. Seed set will then be correlated with pollen stainability scores.

P.1.10 Inheritance studies using $2x$ germplasm (§)

R. Ortiz, D. Vuylsteke and R. Swennen

The inheritance of important traits can be studied preliminarily using $2x$ segregating material already planted at Onne. The traits for evaluation will be BSR, pseudostem color, bunch orientation, dwarfism, apical dominance, parthenocarpy, bunch weight, etc.

P.1.11 Ploidy levels in $3x \times 2x$ hybrids (§)

R. Ortiz, D. Vuylsteke, S. K. Hahn and R. Swennen

Cytological verification of ploidy levels in the progeny from $3x \times 2x$ crosses is needed. Ploidy will initially be estimated by phenotypic appearance and confirmed either by root-tip chromosome counting or DNA flow cytometry analysis. The latter method has been successfully used by TRIP to determine the ploidy levels of cassava and yam cultivars. A protocol for banana/plantain will be developed.

P.1.12 Combining ability in $3x$ plantain germplasm (§)

R. Ortiz and D. Vuylsteke

Progenies from crosses between the French plantain cvs. 'Obino l'Ewal' and 'Bobby Tannap' with the same male parent 'Calcutta 4' differed in ploidy ratio and in the percentage of resistant individuals, with the former cultivar producing a larger number of promising hybrids. Therefore, more detailed assessment of breeding value is needed. Combining ability analysis, using common tester stock(s), will be employed to determine the differences between parents in the production of BSR and high-yielding hybrid progeny.

P.1.13 Production of secondary $3x$ and $4x$ hybrids (§)

R. Ortiz, D. Vuylsteke and R. Swennen

Secondary $3x$ and $4x$ hybrids can be produced by $4x \times 2x$ and $4x \times 4x$ crosses, respectively (Figure 3). High seed set has been obtained in both types of crosses, which challenges the common idea that $4x$ hybrids have low male fertility. Different combinations (2-way, 3-way, and 4-way hybrids) will be compared to determine the effect of heterozygosity on bunch weight and other agronomic traits.

P.1.14 Mid-altitude cooking/beer banana breeding (§)

D. Vuylsteke and R. Ortiz

The highest per capita consumption of *Musa* worldwide occurs in the East African highlands. Cooking and beer bananas, AAA and ABB predominate there. Pest pressure is higher there than in West and Central Africa; *Fusarium* wilt, banana bunchy-top virus, BS and weevil pests are serious biotic constraints. Resistance breeding may be possible since seeds have been produced in a number of these cultivars. The two most common cultivars in that region ('Igitsiri' and 'Igisahira gisanzwe') are multiplied *in vitro* at Onne station to establish a larger pollination block. It is envisaged to send the hybrid materials produced at Onne in seed form to the new IITA Station in Uganda for clonal production, evaluation and selection.

P.2 Breeding for durable sigatoka resistance

Project rationale

Black sigatoka disease (BS), caused by the fungus *Mycosphaerella fijiensis* is considered the major fungal constraint to plantain and banana production worldwide. Introduced to Africa about two decades ago, it spread rapidly through all production zones. It causes severe leaf necrosis and reduces yield by 30-50%. All plantain germplasm at Onne (115 cultivars), collected from West and Central Africa, tropical America and the Philippines is equally susceptible to BS. Chemical controls exist but are economically untenable for the majority of African smallholder farmers, as well as environmentally hazardous. Resistance breeding is generally considered as the most appropriate control strategy. IITA began resistance breeding in 1987. It is important to stress here that a component for the success of the BSR breeding project is the support of PHMD plant pathologists.

Completed studies

Swennen, R. and D. Vuylsteke. 1992. Breeding black sigatoka resistant plantains with a wild banana. (submitted to Tropical Agriculture).

Plantain (*Musa* spp., AAB group), an important staple food crop in the humid tropics of Africa, Latin America and the Caribbean, is threatened by a fungal leaf spot disease called black sigatoka. Two African plantain cultivars of the medium French category were crossed with 'Calcutta 4' (*Musa acuminata* ssp. *burmannicoides*), a wild diploid banana highly resistant to black sigatoka. Four tetraploid hybrids selected from the progenies of the crosses showed high levels of resistance to black sigatoka and produced bunches up to 125% heavier than their fungicide-treated plantain parents. The inferior fruit size of the wild banana was not reflected in these hybrids. The wild diploid 'Calcutta 4' can thus be used in a plantain breeding programme.

Swennen, R. and D. Vuylsteke. 1991. Preliminary results at IITA in breeding plantain for black sigatoka resistance in Africa. In B. Anez, C. Nava, L. Sosa and R. Jaramillo (eds.), Proceedings of the 9th ACORBAT Meeting, held at Merida, Venezuela, 24-29 Sep 1989, pp. 235-244 (1991).

Africa accounts for 50% of all plantain produced in the world. These plantains are overwhelmingly important as a food crop for local consumption. Black sigatoka has now invaded Africa, thereby endangering this important food source. The long term solution to this problem lies in the development of black sigatoka-resistant plantains. The available *Musa* germplasm at IITA was screened for female fertility by pollinating with AA black sigatoka-resistant diploids. Sixteen French plantains and seven False Horn plantains produced seed. Hybrid progenies were rescued using embryo culture techniques. The first black sigatoka-resistant tetraploid plantain hybrids are currently under evaluation in the field.

Vuylsteke, D. and R. Swennen. 1991. Development and performance of hybrids between African plantains and black sigatoka-resistant diploids. Proc. 10th ACORBAT Meeting, Mexico, Nov. 1991. In press.

A strategy to control the black sigatoka disease of plantain in Africa, targeting the incorporation of durable host plant resistance, was initiated at the International Institute of Tropical Agriculture (IITA). The commonly accepted intractability of plantain to genetic improvement has been challenged by the identification of 37 different, seed-fertile plantain cultivars and by the production of 250 hybrids in four years of breeding. Twenty tetraploid hybrids have been selected for their increased black sigatoka resistance, high yields, large parthenocarpic fruits and/or better ratooning. They, however, showed longer bunch maturation times. Seventeen of the selected hybrids had 'Calcutta 4' as paternal source, indicating that the inferior bunch characteristics of this wild banana were not transmitted to its tetraploid progeny. Conversely, the black sigatoka resistance of 'Calcutta 4' was readily inherited in the tetraploid off-spring. Progenies of the triploid

| Step/Activity | Time required |
|--|-----------------------------|
| 1. EVALUATION OF 2x AND 3x GERmplasm 2x: resistance to black sigatoka, good bunch and fruit size, parthenocarpic fruit, good ratooning, dwarfism, male fertility. 3x: female fertility & 2n egg production, combining ability, good fruit/bunch qualities, adaptation to target area. | 1 year |
| 2. a. DIPLOID BREEDING | 1.5 years/cycle |
| b. INTERPLOIDY HYBRIDIZATION: 3x x 2x Utilization of embryo culture for seed germination. | 1.5 years from seed to seed |
| 3. EARLY EVALUATION OF 4x HYBRIDS resistance to black sigatoka and other pests/diseases, earliness, bunch size, good ratooning, fruit parthenocarpy and quality, postharvest storage ability. | 1-2 years |
| 4. SELECTION OF 4x FOR FURTHER EVALUATION AND/OR TO PRODUCE SECONDARY 3x (4x x 2x) or 4x (4x x 4x) hybrids | |
| 5. MULTIPLICATION OF SELECTED CLONES AND REPLICATED TRIALS (Preliminary Yield Trials) Utilization of tissue culture for rapid multiplication | 2 years |
| 6. REGIONAL TRIALS IN WEST, CENTRAL AND EAST AFRICA TO ASSESS YIELD STABILITY AND FIELD RESISTANCE IN COOPERATION WITH NARS | 2-3 years |
| 7. INTERNATIONAL MUSA TESTING PROGRAM Global evaluation of germplasm under auspices of INIBAP | 1 year |
| 8. RELEASE OF ADVANCED GENETIC MATERIALS FOR NARS TESTING AND ON-FARM RESEARCH | |

Figure 1. MUSA BREEDING STRATEGY OF IITA'S PLANTAIN AND BANANA IMPROVEMENT PROGRAM

| STEP | DURATION | TYPE OF TRIAL | NUMBERS INVOLVED |
|------|-------------------------------|--|--|
| 1 | 12-18 months | EET: Early Evaluation Trial (Data recorded on BSR, bunch size, fruit parthenocarpy, dwarfness) | <u>≥ 100 clones</u> 1-5 plants/1 rep. |
| | | ↓ | |
| 2 | 1-2 years | PYT: Preliminary Yield Trial (Data recorded idem step 1 + earliness, ratooning, postharvest quality & durability, and other pest/disease resistance) | <u>25-30 EET selected clones</u> RCBD or lattices with 2 reps. of 4 plants each |
| | | ↓ | |
| 3a | 2 years BSR stability) | MET: Multilocational Evaluation Trial (Data recorded to assess yield and | <u>8-15 PYT selected clones</u> + parents & local cvs as checks. RCBD using 5 plants/ 2 reps/5-15 locs. |
| | | ↓ | |
| | | 3b Other breeding materials coming from conventional or biotech programs through global network testing (IMTP/INIBAP) | |
| | | ↓ ↓ | |
| 4 | 2-3 years | AMYT: Advanced Musa Yield Trial (Data recorded idem 3 + standards of local preferences of each NARS. NARS to supply local cultivars) | <u>3-5 advanced selected MET/IMTP clones</u> RCBD using 5 plants/ 2-4 reps/ ? locs. |
| | | ↓ | |
| 5 | # years | OFT: On-Farm Trials (to release new cultivars & develop agronomic practices to maximize its yield: "cultivar profile") | <u>1-2 new cvs.</u> with traditional cvs. # plants, # reps, # treatments (density, mulching, N-P-K, etc.) depending on NARS |

Steps 1 & 2 done at Onne (Nigeria) by IITA/PBIP
Step 3a done by IITA in collaboration with NARS
Step 3b done by INIBAP in collaboration with IITA & other breeding programs and NARS
Step 4 done by NARS with IITA & INIBAP support
Step 5 done by NARS (if required with inputs from IITA & INIBAP)

Figure 2. PROPOSED FLOW OF IITA MUSA BREEDING MATERIALS IN AFRICA

French plantain cvs. 'Obino l'Ewai' and 'Bobby Tannap' differed in their black sigatoka breeding values, the former producing larger numbers of promising hybrids. Tetraploids produced from crosses of plantain cultivars with the homozygous 'Calcutta 4' displayed variation in black sigatoka reaction, phenotype and growth and yield parameters, suggesting the occurrence of recombination in the fully restituted, triploid plantain genome during modified meiosis.

Pasberg-Gauhl, C., F. Gauhl and D. Vuylsteke. 1991. Sigatoka research in the Plantain and Banana Improvement Program (PBIP) at IITA, Nigeria. Proc. 10th ACORBAT Meeting, Mexico, Nov. 1991. In press.

About 15 years ago, the black sigatoka (BS) disease, caused by the fungus *Mycosphaerella fijiensis*, was accidentally introduced to Africa. No resistance or tolerance to the disease has been found among the 115 plantain (*Musa* AAB) cultivars in IITA's collection. In 1987 a program aimed at breeding for durable host plant resistance was initiated. Several promising hybrids have been produced. A clonal evaluation trial of IITA hybrids was started in October 1990. In August 1991 a multilocal trial of 18 different *Musa* clones, including 8 IITA hybrids and 10 reference clones started. Additional research involves the understanding of the epidemiology and biology of *M. fijiensis*. This includes studies of ascospore release and dispersal. A volumetric spore trap with an automatic data logging weather station was recently set up for detailed ecopathology analysis in the humid forest zone of West Africa.

F. Gauhl, K. N. Mobambo, C. Pasberg-Gauhl, R. Swennen and D. Vuylsteke. 1991. Preliminary evaluation of black sigatoka resistance in IITA plantain hybrids. Proc. 10th ACORBAT Meeting, Mexico, Nov. 1991. In press.

A clonal evaluation trial of IITA plantain hybrids for black sigatoka (BS) resistance was established in October 1990. The trial includes the three tetraploid hybrids TMPx 548-4, TMPx 548-9, and TMPx 597-4, and their non-treated and fungicide-treated female plantain parent 'Obino l'Ewai' (French plantain). Differences between the BS susceptible plantain parent and the hybrids were distinct. In the first week of April 1991 (6 MAP) the number of standing leaves in the non-treated plantain was 10.0, in the fungicide-treated plantain 12.9, whereas TMPx 548-4 and TMPx 548-9 had 11.4 and TMPx 597-4 had 11.9 leaves. The youngest leaf spotted (spot with dry center) in the non-treated plantain was 5.5, in the fungicide-treated plantain 6.6, in TMPx 548-4 and TMPx 548-9 11.2 and in TMPx 597-4 9.9. The percentage of infested leaf area was 21.5, 18.3, 2.2, 2.1 and 2.9, respectively. For the parameter "youngest leaf with symptoms", the values ranged between 2.0 and 2.5.

Activities

P.2.1 Generating BSR 4x hybrids of plantains

D. Vuylsteke, R. Ortiz and R. Swennen

Currently, PBIP focuses on the production of BS resistant (BSR) 4x hybrids of plantains through 3x x 2x crosses. About 10,000 seeds are produced annually from crosses of plantains with different diploids. Of about 500 plantain hybrids germinated *in vitro* and transferred to the field nursery, 250 were eliminated early on because of grossly abnormal foliage typical of aneuploids and higher polyploids. The clone 'Calcutta 4' (*M. acuminata* ssp. *burmannicoides*) has been the most frequently used 2x male parent in the production of BSR hybrids. This clone has a high level of BS resistance and readily transmits this trait to its offspring. The degree of BS susceptibility varied among the hybrid 4x progeny. Twenty 4x hybrids of plantain have been selected that combine partial to high resistance to BS with good bunch and fruit characteristics, resulting in high yield (ranging from 11.5 to 21.8 kg per bunch against 12.3 to 14.0 kg for the plantain parents). Three of the BSR plantain hybrids have been planted in a clonal evaluation trial at Onne Station. Two BSR cooking banana hybrids have also been selected. Additional BSR material comprised 4 East African highland banana hybrids.

P.2.2 Field evaluation of BSR 4x hybrids at Onne

D. Vuylsteke, R. Ortiz, F. Gauhl,

C. Pasberg-Gauhl, K. Mobambo and R. Swennen

This experiment (with RCB design) is carried out to evaluate BSR and yield performance of tetraploid hybrids as compared to the maternal (plantain) cultivar under fungicide and non-fungicide conditions at Onne. Yield loss due to black sigatoka disease was also assessed. Plantain yield loss due to black sigatoka disease was 33%, as estimated from the difference in yield between the fungicide-treated and non-treated 'Obino l'Ewai' (23.5 and 15.8 t/ha, respectively). Preliminary results of BS scoring indicated significantly less leaf spot damage in the 3 tested hybrids (TMPx 548-4, 548-9 and 597-4) as compared to the plantain cv. 'Obino l'Ewai'

from which they were derived. Yield in the plant crop of hybrid TMPx 548-9 was 43% higher than that of the fungicide-treated and twice that of the non-treated 'Obino l'Ewai' (33.5, 23.5 and 15.8 mt.ha⁻¹, resp.). This suggests that the higher yield of TMPx 548-9 was not only due to its BS resistance, but also to heterosis. Hybrid TMPx 597-4, although showing low BS disease scores in the vegetative growth stage, had poor yield performance (12.7 mt.ha⁻¹). This suggests that tetraploidy *per se* does not always increase yield. Hybrid TMPx 548-4, which also showed good resistance, performed better (23.8 mt.ha⁻¹) than the non-treated plantain, but similar to the fungicide-treated 'Obino l'Ewai'.

P.2.3 Multi-site evaluation of BSR 4x hybrids

D. Vuylsteke, R. Ortiz, F. Gauhl and C. Pasberg-Gauhl

The objective of this set of replicated trials is to test the performance of the IITA improved BSR 4x plantain hybrids under different agroecological conditions. A first set of BSR 4x hybrids (seven plantain- and one cooking banana-derived) are currently undergoing multilocational evaluation trials (MET) in Nigeria and Cameroon. Three BSR 3x cooking banana cultivars and 7 reference clones (including the 3x and 2x parents of the hybrids) have also been included in this first MET, which was planted in August and September 1991. The importance of G x E interaction and the stability of the different genetic materials in terms of yield and BSR will be determined, and the symptomatology and pathogenic variation of BS in different West and Central African environments will be analyzed.

P.2.4 Inheritance of black sigatoka resistance (§)

R. Ortiz and D. Vuylsteke

This activity will focus on the study of the inheritance of black sigatoka resistance using 2x material to facilitate the genetic analysis. The genetic nature of BSR will be determined by using different mating designs (parent-offspring, hierarchical design, factorial design and generation mean analysis) and biometrical methods currently available. The parent-offspring regression analysis will be done with material already planted at Onne. The hierarchical and factorial mating designs will be completed during 1992 and the experiments planted in 1993. Generation mean analysis requires the identification of homozygous susceptible and resistant parents. Potential candidates are *M. acuminata* ssp. *banksii* (237) and 'Calcutta 4' (*M. acuminata* ssp. *burmannicoides*). These clones will be crossed to produce F1 progeny during 1992. Next year the F1 will be selfed and backcrossed to each parental line to complete the generations required for the analysis.

P.2.5 Multilocational evaluation trials (MET) (§)

R. Ortiz, D. Vuylsteke, F. Gauhl and C. Pasberg-Gauhl

The objective of this series of trials is to investigate G x E interaction for important traits and to determine the yield and BSR stability of promising hybrids. These trials are thus an advanced testing stage of the breeding scheme (see Figs. 1 and 2). The performance of twelve 4x BSR hybrids, along with their three female 3x parents ('Obino l'Ewai', 'Bobby Tannap' and 'Bluggoe'), two male 2x parents ('Calcutta 4' and 'Pisang lilin') and three reference cultivars ('Agbagba', 'Cardaba' and 'Valery'), will be evaluated in the second MET. The proposed testing sites are the IITA-Onne station, NIHORT and other locations in Nigeria, the CRBP station at Njombe and the IITA station at Mbalmayo in Cameroon, and CRI in Ghana. A strong research collaboration with NARS and CRBP is anticipated.

P.2.6 International Musa Testing Program (§)

D. Vuylsteke, R. Ortiz, F. Gauhl, C. Pasberg-Gauhl and INIBAP and FHIA scientists

The IMTP, set up by INIBAP, aims at the identification of germplasm (natural or improved) with resistance/tolerance to a specific disease or pest through testing on a global scale. The first IMTP concentrates on black sigatoka resistance and involves 6 selection sites in two continents (Africa and tropical America). IITA participates as an equal partner in the IMTP being carried out in Africa. The current IMTP-1 trial centers on the evaluation of 7 hybrids of banana and plantain produced by FHIA (Fundacion Hondurena de Investigacion Agricola, La Lima, Honduras), along with 11 BS standard hosts and reference cultivars in observation (unreplicated) plots. This research is part of IITA's collaboration with INIBAP and FHIA. In the near future, IITA BSR hybrids will be ready to enter the IMTP-1.

P.3 Biotechnology for *Musa* breeding

Project rationale

Efforts to propagate, conserve and breed cultivated *Musa* are fraught with technical obstacles specific to the biology of this vegetatively propagated crop such as slow propagation, low fertility in crosses and limited genetic variability. An array of tissue culture and molecular genetic techniques show promise to facilitate handling and improvement of *Musa* germplasm.

Completed studies

Vuylsteke, D., J. Schoofs, R. Swennen, G. Adejare, M. Ayodele and E. De Langhe. 1990. Shoot-tip culture and third-country quarantine to facilitate the introduction of new *Musa* germplasm into West Africa. *Newsl. FAO/IBPGR Plant Genetic Res.* 81/82: 5-11.

Tissue culture is increasingly being used as a vehicle for the safe exchange of *Musa* germplasm. However, certain pathogens, e.g. banana bunchy-top virus (BBTV), may pass undetected through *in vitro* culture. To meet the urgent need for introducing new desired germplasm for *Musa* breeding and reduce the risk of pathogen transfer, a germplasm-movement strategy based on two independent safeguards, viz. shoot-tip culture and third-country quarantine, was adopted. More than 270 *Musa* accessions have been introduced into Nigeria, including 33 with black sigatoka resistance. BBTV has not been diagnosed in any of the introduced materials.

Vuylsteke, D., R. Swennen and E. De Langhe. 1991. Somaclonal variation in plantains (*Musa* spp, AAB group) derived from shoot-tip culture. *Fruits* 46(4): 429-439.

The nature and extent of somaclonal variation in micropropagated plantains (*Musa* spp, AAB group), and factors influencing its incidence, were investigated. Plantain cultivars varied widely in terms of their *in vitro* stability as determined by the occurrence of variation in the phenotype of regenerated plants. Different cultivars also differed in the characters affected by phenotypic variation. Much of the somaclonal variation mimicked the variability occurring naturally, in that the variants of a cultivar were essentially copies of other plantain cultivars. Traits affected by phenotypic change were inflorescence morphology and associated female fertility, fruit shape, pseudostem, petiole and bract colour, and plant stature. Abnormal foliage was observed in some cultivars. Many of the phenotypic variants were stable through several cycles of vegetative propagation, suggesting that much of the variation was genetic in origin. Somaclonal variation was not correlated with time in culture. There was a trend of higher variation frequencies with increasing *in vitro* proliferation rates. Analysis of source identity of regenerated plants indicated that variation arose in culture, suggesting that the tissue culture environment imposed a stress that induced the changes. The narrow spectrum of somaclonal variation was of limited use in plantain improvement. Through its increased seed fertility, the French inflorescence-type variant may be of value in breeding the preferred, but highly sterile False Horn plantain, from which it was derived.

Swennen, R., D. Vuylsteke and S.K. Hahn. 1990. The use of a few simple biotechnological tools in wide crosses involving plantains. *Proc. Italy/CTA/FAO/IITA Conference on Biotechnology. IITA, November 1990 (In press).*

Plantains are a staple food crop in sub-Saharan Africa. Black sigatoka, a leaf spot disease, is the principal cause of current yield reductions. A plantain breeding program was mounted at IITA which produced four black sigatoka-resistant plantain hybrids nearly three years after its inception by crossing the triploid plantains with a wild diploid. A few simple biotechnological aids, such as introduction of germplasm through *in vitro* meristem cultures, rapid micropropagation and embryo-rescue culture, were pivotal in this speedy achievement.

Vuylsteke, D. and R. Swennen. 1990. Biotechnological approaches to plantain and banana improvement at IITA. *Proc. Italy/CTA/FAO/IITA Conference on Biotechnology. IITA, November 1990 (In press).*

The potential of biotechnologies in banana and plantain improvement are considerable because *Musa* spp. are intractable in terms of conventional breeding strategies. Simple tissue culture techniques, such as shoot-tip culture and embryo culture, can overcome some obstacles that impede breeding progress. These have contributed to IITA's success in the production of black sigatoka-resistant plantains. Assistance in the development of tissue culture laboratories has been provided to Nigerian national programs, thereby strengthening their capability in biotechnology. Cell culture and RFLP mapping, both essential to the future implementation of molecular genetic techniques in *Musa* improvement, are being researched in collaboration with advanced laboratories.

Dhed'a, D., F. Dumortier, B. Panis, D. Vuylsteke and E. De Langhe. 1991. Plant regeneration in cell suspension cultures of the cooking banana cv. 'Bluggoe' (*Musa* spp. ABB group). *Fruits* 46(2):125-135.

Embryogenic cell suspension cultures of the widespread cooking banana clone 'Bluggoe' (*Musa* spp., ABB group) were established by culturing meristematic «scalps», taken from proliferating shoot-tip cultures, in a modified liquid MS medium containing 5 µM 2,4-D and 1 µM zeatin. Plant regeneration from suspension cultures was achieved through a sequence of four steps involving different media. Embryogenic globules formed when a sieved suspension was cultured in liquid medium without growth regulators. The addition of cytokinin was essential for the subsequent maturation and germination of these globules and resulted in plant recovery frequencies of 10-14.5%. Plant regeneration proceeded through the developmental pathway of somatic embryogeny, which at all stages showed conspicuous morphological and histological resemblance with zygotic embryogenesis in a wild *Musa* species. Somatic embryos were produced directly from cells in suspension and not via callus. Germinated banana somatic embryos were successfully established in soil. The relative simplicity of this cell culture protocol may enhance the feasibility of integrating biotechnological approaches in conventional schemes of banana and plantain improvement.

Activities

P.3.1 *Musa* germplasm exchange *in vitro*

D. Vuylsteke and S. Ng

Aseptic shoot-tip culture, in combination with third-country quarantine, is used for the safe exchange of banana/plantain germplasm. Shoot-tip cultures confer considerable advantages: (1) the mass of plant material moved is greatly reduced, (2) the plant material is contained in a sealed vessel, (3) they are free of pests and pathogens, and (4) they are amenable to rapid multiplication. From 1985 to 1990, 320 new accessions were introduced by IITA as *in vitro* cultures, thereby quadrupling the number of accessions held in the collection. In 1991, 28 new germplasm accessions were introduced from 5 countries, mainly Colombia and Honduras. These genetic resources, which provide the basis for the breeding program, were introduced in a joint effort with the Nigerian Plant Quarantine Service and the INIBAP Transit Center at K.U. Leuven (Belgium). IITA also deposited 18 hybrids of banana and plantain at the INIBAP Transit Center for virus indexing in view of their possible future international dissemination. Over 500 germplasm accessions, among which are 110 plantain cultivars and about 300 plantain hybrids, are maintained *in vitro* for germplasm conservation and as duplicates of the field genebank (see xxx). The *in vitro* collection is partly duplicated at the Ibadan tissue culture laboratory for safety reasons, where it is maintained under minimal growth conditions.

P.3.2 *In vitro* propagation of selected genotypes

D. Vuylsteke

Shoot-tip culture is a well-established, adequate and relatively simple *in vitro* method for the rapid propagation of *Musa* and the production of disease-free planting material. Multiplication rates are several orders of magnitude higher than those obtained with conventional methods, which enables rapid multiplication of newly-bred genotypes for testing or release. Over 500 *Musa* accessions of seven different genomic constitutions and ploidies have been successfully propagated *in vitro* at PBIP's tissue culture laboratory at the Onne Station. Micropropagation has been pivotal in the rapid deployment of the breeding program by supplying large numbers of plants of female and male parents for the crossing blocks and of BSR hybrids for evaluation trials. About 4,000 plants were produced for the latter purpose in 1991. Micropropagated plants of BSR cooking bananas (*Musa* AAB and ABB) were distributed to Nigerian national programs through an INIBAP-supported activity. In 1991, 4200 plants of cvs. 'Muracho', 'Fougamou', 'Pelipita' and 'Cardaba' were distributed in this scheme. Technical support on *in vitro* propagation of *Musa* was provided to two tissue culture laboratories of Nigerian NARS.

P.3.3 Embryo rescue in *Musa*

G. I. Harry, D. Vuylsteke and U. U. Eborg (Rivers State Univ. of Science and Technology)

Hybrid plant production in the most common triploid *Musa* clones is hampered by low seed set and low seed germination rates. Seeds of plantain crosses germinate in soil at a rate of only 1%. Aseptic embryo culture techniques are routinely applied to increase seed germination rates by a factor of 3 to 10. An average of about 900 plantain hybrid seeds are handled *in vitro* on a monthly basis in support of the breeding activity. A research fellow is investigating the possibility of rescuing immature embryos to further enhance germination rates. Preliminary results showed that immature embryos 50-55 days after pollination had 20.7% germination as compared to the 1.0% germination of mature embryos (80-85 days after pollination).

P.3.4 Somaclonal variation studies in *Musa*

D. Vuylsteke, R. Swennen, D. Dhed'a, B. Panis

The increased genetic variation among plants regenerated from *in vitro* culture has been termed as somaclonal variation, a ubiquitous phenomenon. The frequent use of *in vitro* culture techniques for the handling of *Musa* germplasm warrants investigations into the occurrence of somaclonal variation in this genus. Somaclonal variation in plantain plants derived from shoot-tip culture is described in detail elsewhere (see Completed studies). Within the homogeneous plantain subgroup, the rate of variation was cultivar-specific, ranging from 0 to 69%. In collaboration with the K.U. Leuven (Belgium), the genetic stability of cooking banana plants (*Musa* spp. ABB group, cv. 'Bluggoe') regenerated from cell suspension cultures by somatic embryogenesis is being investigated. Only 1 plant among 140 regenerants so far showed aberrant leaf morphology, the remaining plants being phenotypically normal. Cryopreservation of cell suspension cultures of the cooking banana cv. 'Bluggoe', and plant regeneration from single cells was recently achieved at K.U. Leuven. In a continued joint effort, plants obtained from such cryopreserved cell suspensions are screened for somaclonal variation at Onne station. This activity will help in assessing the problems and potentials of long term conservation of *Musa* and other crops, through cryopreservation of *in vitro* cultures.

P.3.5 Causes of somaclonal variation in *Musa* (§)

D. Vuylsteke, L. Withers, R. Swennen, R. Ortiz and possibly other collaborators

Somaclonal variation can undesirably alter the genetic makeup of improved germplasm propagated *in vitro*. Many somaclonal variants are not apparent until in the field, often only at fruiting. This has serious practical and economic consequences for banana/plantain production at the farm level, threatens the security of *in vitro* conservation, and hampers the efficient application of biotechnological approaches to genetic improvement. A project under FAO's Associate Professional Officer (APO) scheme in collaboration with IBPGR, K.U. Leuven and others is proposed to further our understanding of this problematic phenomenon. The top priority objective is to devise improved *in vitro* culture management procedures for the control of genetic instability. The project will also explore methods for the early detection and characterization of somaclonal variants using biochemical/molecular markers. This endeavour may result in a genetic mapping of the genes involved in somaclonal variation in *Musa*, which could elucidate the underlying causes and origins of this phenomenon.

P.3.6 Molecular methods in *Musa* breeding

R. Jarret, D. Vuylsteke and R. Ortiz

This is a collaborative project with Dr. R. L. Jarret (USDA/ARS, Georgia, USA). Interspecific and intersubspecific relationships between the different *Musas* were investigated using RFLP. The relationships between most species were in agreement with conventional morphology-based phylogeny with few exceptions: *M. baccarii* and *M. basjoo*. The degree of polymorphism in 10 plantain (AAB) cultivars was studied using 5 restriction enzymes x 20 low copy number sequences (=100 probe enzyme combinations). No polymorphisms were detected. In a similar way, the discriminating ability of random amplified polymorphic DNA (RAPDs) using commercially available random primers was evaluated. Additional primers were obtained from a 520 bp highly-repetitive and dispersed sequence originally isolated from the *M. acuminata* ssp. *malaccensis* genomic library. This sequence is extremely AT enriched. PCR amplification of the synthesized primers was able to detect polymorphism among the cultivars examined. Therefore, RAPDs seem to be quite effective in detecting polymorphisms, especially when used in conjunction with one of the synthesized primer sequences internal to the 520 bp repeated sequence. Another objective of the project is to differentiate the somaclonal variants arising from tissue culture from their normal parents using molecular markers (RFLPs, RAPDs and VNTRs). Initial attempts to isolate DNA from leaf tissue collected at Onne has been unsuccessful. In order to have proper leaf material it must be harvested at a very young stage of development (prior to leaf expansion), frozen quickly and lyophilized for 72 hours. For this activity a small freeze drier needs to be acquired at Onne Station.

P.3.7 Developing a molecular linkage map in *Musa* (§)

R. Ortiz, D. Vuylsteke and R. Jarret

The availability of molecular markers (MM, e.g., RFLPs, RAPDs, VNTRs) in *Musa* provides the opportunity to develop a linkage map. Two diploid segregating populations of *M. acuminata* (AA) developed using a homozygous and two heterozygous parents will be used, in collaboration with Dr. R.L. Jarret (USDA/ARS). The materials are already planted at Onne. The segregating populations are genetically equivalent to test crosses. Thus, two point test cross analysis will be used to determine the recombination fraction between markers. The linkage map will then be developed using available software (either LINKAGE@ or MAPMAKER@). The ability to accurately estimate genetic relatedness in germplasm collections will be an

important tool in the conservation and management of the genetic diversity in genebanks. The inability to estimate genetic diversity results in a risk that unique materials may not be added to the collection or potentially valuable materials may be discarded. MM are important new tools that can "fingerprint" accessions. MM are potentially useful not only to estimate genetic diversity but also to characterize somaclonal variants of *Musa* propagated *in vitro*. In this way, both genetic identity and stability can possibly be defined. MM can also be used to determine the origin (hybrids vs. parthenogenetic derived) of the 4x material produced from 3x x 2x crosses.

P.3.8 Using molecular markers to map BSR (§)

R. Ortiz, D. Vuylsteke and R. Jarret

This activity will be in collaboration with Dr. R.L. Jarret (USDA/ARS). The populations described in P.2.4 (generation mean analysis) will be used for this activity. The objective is to identify molecular markers linked to BSR loci. In this way, it is expected that quantitative trait variation (QTV) for BSR can be dissected into its components and the main types of gene action controlling BSR phenotypic expression can be assessed. Furthermore, the markers linked to BSR could be useful for marker assisted selection (MAS) in the development of 2x, 3x and 4x breeding materials during the early stages of plant development.

P.4 Postharvest quality of plantains

Project rationale

Promising new BSR hybrids arose from crosses with wild relatives, which have much different fruit characteristics. Hence, hybrids are likely to have altered fruit quality which will probably affect consumer acceptability. Research is needed to determine the criteria associated with plantain fruit quality and to give breeders methods for rapid, efficient screening to identify superior clones.

Completed studies

Eggleston, G., R. Swennen and D. Vuylsteke (unpubl.). A survey of consumer reaction to BSR hybrids. A preliminary survey has been conducted to clarify any taste differences between plantain cultivars. This information could be an aid in the selection of plantain hybrids. An initial study in consumer acceptability revealed no difference in taste between the BSR hybrid TMPx 548-9 and its susceptible plantain parent 'Obino l'Ewai'. However, fruit color of the hybrid was less preferred. Therefore, 'Obino l'Ewai' had significantly better consumer acceptability than the BSR hybrid, which was, however, still rated as good.

Eggleston, G., R. Swennen and S. Akoni. 1992. Physicochemical studies on starches isolated from plantain cultivars, plantain hybrids and cooking bananas. Starch/Stärke (In press).

Starches from mature, unripe fruit pulp of plantain cultivars (*Musa* spp., AAB group) representing the wide variability in African tetraploid and diploid plantain hybrids and starchy cooking bananas (*Musa* spp., ABB group) were isolated and characterised. In general, studies revealed very compact irregularly shaped and sized granules, with low amylose content (9.11 - 17.16%), highly resistant to bacterial α -amylase attack. Brabender amylograms showed very restricted swelling type patterns with great stability and negligible retrogradation. Results indicate that differences in physico-chemical properties exist amongst the three *Musa* fruit group starches. Plantains represent a chemical/molecular homogenous group, but are heterogeneous for granule structure. Ploidy level affected hybrid properties. ABB cooking bananas exhibited highly pronounced restricted swelling and high gelatinisation and pasting temperatures, indicating a more ordered, very strongly bonded granule structure; chemical and physical properties varied considerably within the ABB genotype.

Activities

P.4.1 Fruit quality in plantain/cooking banana (§)

S. Ferris, R. Ortiz and D. Vuylsteke

This postdoctoral project will investigate fruit palatability (flavour, texture, response to cooking procedures) and durability (shelf life, ripening, handling and storage aspects) with the goal of defining the important quality parameters and identifying rapid screening techniques for breeders' use. The methodologies involved are sensory evaluation (taste panels), surveys and physicochemical measurements.

P.5 Sustainable perennial plantain production

Project rationale

Careful management of organic matter is essential to achieve sustained perennial productivity of plantain under large scale field production conditions. Agroforestry systems such as alley cropping and the management of regrowth of natural bush fallow species in plantain fields are being investigated for their capacity to maintain productivity over long periods of cultivation without degradation of the resource base.

Activities

P.5.1 The perennial yield potential of plantains

D. Vuylsteke, R. Swennen and S. Witters

This experiment, established in 1987, will continue for at least 5 years. It has several objectives: a) to determine the yield potential of 6 cultivars representative of cultivar variability in the plantain subgroup over time, b) screen for somaclonal variation among *in vitro* propagated plantains on a wide genotypic basis, and c) study sigatoka epidemiology and assess yield loss over a longer time period. The varieties included are the French plantains 'Ntanga 2', 'Bobby Tannap', and 'Obino l'Ewai', the False Horn plantains 'Big Ebanga' and 'Agbagba', and the Horn plantain 'Ubok lba'. Other treatments are replanting vs. normal ratooning after harvest. Results from the plant crop indicated large differences in yield among some cultivars. The French plantain cvs. 'Ntanga 2' and 'Bobby Tannap' were highest-yielding (21.5 and 19.9 t/ha-yr, resp.), the French cv. 'Obino l'Ewai' and the False Horn cvs. 'Big Ebanga' and 'Agbagba' were intermediate (16.7, 14.5 and 14.0, resp.), and the Horn plantain cv. 'Ubok lba' had the lowest yield (8.8 t/ha-yr). After adjusting the yields to a per unit time basis (t/ha-yr), differences were still evident between the three inflorescence-type categories, but not within categories (except for 'Obino l'Ewai' within the French plantains).

P.5.2 Alley cropping with plantain

B. Ruhigwa, R. Swennen and M. Gichuru

A Research Fellow from Zaire carries out this research activity at Onne station to a) develop a more efficient production system for small farmers, b) test tree leaves and litter as mulch for plantain, and c) test different tree species for mulch production under shade of plantains. He found that plantains in *Acioa barterii* alley cropping plots and in elephant grass (*Pennisetum purpureum*)-mulched plots performed better than in treatments in which other hedgerow species (*Gmelina arborea*, *Alchornea cordifolia*, *Cassia siamea*) were used. The plantain yields were slightly lower in the *Acioa* alleys than in the control *Pennisetum* mulch plots (15.1 and 17.8 mt.ha⁻¹, resp.); however, the *Pennisetum* system requires higher land and labour inputs because mulch must be transported from external fields. Therefore, alley cropping with *Acioa* is a more promising system for field production of plantains. This research has been carried out with the cooperation of the Resource and Crop Management Division.

P.6 NARS collaboration and training

Project rationale

Because of the very recent advent of IITA's plantain/banana improvement work, NARS are less developed than for other IITA crops. PBIP acts to strengthen NARS through a) cooperative research, b) consultancy support, c) individual training: scientific visits and Ph.D. students, d) group training, and e) transfer of advanced *Musa* genetic materials and production packages.

Completed studies

Vuylsteke, D. and R. Swennen (unpubl.). Transfer of tissue culture technology to NARS.

Technical support on *in vitro* multiplication techniques was provided to several NARS. Imo State Agricultural Development Project at Owerri, Nigeria was assisted in establishing a tissue culture laboratory in 1989 which now uses the technique for rapid multiplication and distribution of BSR *Musa* to farmers.

Swennen, R. 1990. Plantain cultivation in West African conditions: a reference manual. IITA, Ibadan, Nigeria. 24 p.

This manual summarizes key findings from 10 years of physiological and agronomic research at IITA. It has been distributed to NARS, trainees and farmers to help improve plantain cultivation.

Vuylsteke, D. 1989. Shoot tip culture for the propagation, conservation and exchange of *Musa* germplasm. IBPGR, Rome.

Practical manual for handling crop germplasm *in vitro*. A comprehensive view of current *in vitro* techniques for *Musa* germplasm conservation and exchange.

Activities

P.6.1 International Musa Testing Program (§)

See P.2.6.

P.6.2 Multilocation evaluation trials (§)

R. Ortiz, D. Vuylsteke, F. Gauhl and C. Pasberg-Gauhl

Refer to P.2.5. The Nigerian Interministerial Committee for Black Sigatoka has required the concurrence of PBIP for the development of new breeding materials and technology in this area. Seven Nigerian sites will be included in the MET-2 for this purpose.

P.6.3 Cooking bananas for Nigeria (§)

D. Vuylsteke

PBIP cooperation has been required by the Commissioner of Agriculture of Rivers State, Nigeria for launching the cooking banana "Cardaba" as new potential cultivar for this area. PBIP will be providing planting materials (suckers) for further distribution to farmers by the government.

P.6.4 Graduate training

1. *In vitro* embryo rescue for the enhancement of hybrid plantain production - G. I. Harry (Ph.D. thesis), D. Vuylsteke and U. U. Eborg (Rivers State Univ. of Science and Technology). See P.3.3.
2. Growth parameters of yield in the plant and ratoon crop of six cultivars representing wide variability in plantain - S. Witters (tr. thesis), D. Vuylsteke and R. Swennen (KUL, Belgium). See P.5.1.

P.6.5 Plantain training course (§)

D. Vuylsteke, R. Ortiz and other IITA and NARS scientists

This annual course, "Plantain production and technology transfer", is offered for three weeks in November to young African scientists and technicians. NARS scientists participate as resource persons on specific topics.

Genetic Resources Unit

The Genetic Resources Unit (GRU) has the responsibility to collect, characterize, conserve and distribute the germplasm of food legumes, rice, root and tuber and plantain/banana crops in Africa. Maize may be added to this list as no IARC is currently conserving African traditional varieties. Small collections of a few other crop and agroforestry species are also held to service particular needs of Program researchers and researchers from other IARCs such as ICRAF. The existing field collection of cassava germplasm previously held by the Root and Tuber Improvement Program was transferred to GRU in 1991 and all the germplasm materials are now consolidated and maintained by the Unit. Plantain and banana germplasm is mainly an active working collection, kept at the Onne Station where researchers are located who manage the material. These collections are essential tools for IITA's breeding programs, as well as for researchers world-wide and for posterity.

G.1 Germplasm collection

Project rationale

Collection of the germplasm African food crops is essential to partially counteract the genetic erosion that is occurring as a result of development on the continent. We organize collections in-country with NARS on a targeted basis. This germplasm serves as an invaluable resource for future crop improvement efforts.

Completed studies

Ng, N.Q. and S. Padulosi (unpubl.). Previous plant exploration and collection at IITA.

During 1985-90, the Unit mounted 18 exploration missions to 17 African countries. Exploration is done in close collaboration with NARS; we provided technical and financial support to the national programmes in the Republic of Benin and Ghana for exploration in their countries. Special-funded (Government of Italy) explorations for cowpea (*Vigna unguiculata*) and its wild relatives began in 1985 and for root and tuber germplasm in 1989. Expansion of the gene pool of cowpea through collection of wild *Vigna* is a priority because high levels of insect resistance, the major production constraint for the crop, are not found within the cultivated species. The Unit has also contracted collaborators from the Plant Germplasm Institute, Bari, Italy to explore and collect wild *Vigna* from South Africa and Namibia and cultivated cowpea germplasm from the Mediterranean region. Since 1988, high priority has also been given to yam. IITA's other crops are also collected in the course of *Vigna* and yam exploration missions. Although no exploration was carried out directly by the Unit during 1991, our collaborators explored in Ghana and Namibia. The Unit continues to receive germplasm materials collected in earlier missions in the Republic of Benin, Ghana and Namibia and from miscellaneous collaborators elsewhere.

Ng, N.Q., R. Asledu and S.K. Hahn (unpubl.). Survey of the status of cassava germplasm collection in Africa.

In early 1991, questionnaires for the survey of the status of cassava germplasm collection in Africa were sent to 32 national scientists and authorities in major cassava growing countries in Africa. Results indicate that approximately 3,500 accessions of local cassava are held in 16 African countries. Almost all countries surveyed indicated a need for further collection as well as characterization of their local germplasm. At the meeting of collaborators in Root and Tuber Crop Improvement and Production System Research held at IITA June 11-13, 1991, participants confirmed this need and stressed the need for training NARS researchers in the relevant skills, including *in vitro* culture for storage and disease-free dissemination. They also recommended that a duplicate of the existing NARS cassava collections be conserved at IITA.

Activities

G.1.1 Collection of cassava in Nigeria (§)

N.Q. Ng, F.I. Nweke and Nigerian NARS

COSCA village interviews indicate that 1,000 locally named cassava varieties are being grown in the six African countries surveyed (Cote d'Ivoire, Ghana, Nigeria, Tanzania, Uganda and Zaire). Plans have been made with COSCA to collect and assemble this germplasm. GRU will join the COSCA team in March-April 1992 to specifically collect the named cassava cuttings in Nigeria when the team revisits farmers they interviewed previously. Collections in other countries will also be arranged.

G.1.2 Collection in Congo and Uganda (§)

S. Padulosi

Uganda has not yet been explored by the Unit. Congo is an area with high diversity for wild cowpea (*V. unguiculata*) and Uganda has among the largest number of wild *Vigna* species in Africa. Germplasm of IITA's other mandated crops will also be collected.

G.1.3 Collection in Guinea-Conakry (§)

N.Q. Ng, F.L. Guilavogui, El Sanonssy Bay and M.D. Winslow

Funded by the RRPMP project and based on a request by the national program, tentative plans have been made to explore and collect cassava and maize germplasm in Guinea (Conakry) in late 1992. A member of the Unit or a short-term consultant will lead the exploration team with two national scientists. Other crops and their wild relatives, particularly *Vigna* and *Dioscorea* will also be collected.

G.2 Germplasm conservation and distribution

Project rationale

Germplasm conservation is a high priority worldwide, and crop improvement researchers have a special responsibility to conserve germplasm since the successful spread of improved varieties is one cause of the erosion of existing genetic diversity in crop species. Unrestricted availability of this germplasm is a guiding principle of the CGIAR which the Unit implements.

Activities

G.2.1 Cowpea germplasm conservation

N.Q. Ng, A.E. Adegbite, J. Obarinde and J. Apeji

In 1991, 3,880 of the total 15,185 cowpea accessions were multiplied/rejuvenated, mostly during the second and dry seasons at Ibadan. All are maintained in an active collection store (5°C, 30% RH). About 7,500 accessions are also in long-term storage in sealed aluminium containers (-20°C, 5% seed moisture content). About 272 new accessions were characterized for up to 30 traits, with data stored in a computer data bank.

G.2.2 Cowpea core germplasm set (§)

N.Q. Ng

Data on agrobotanical traits, resistance to diseases, pests, and physiological stresses as well as agroecological adaptation will be analyzed to choose accessions for a core collection sampling the range of diversity for useful traits in cowpea. If necessary for more detailed discrimination, additional biochemical data will be gathered or generated. This core collection will be valuable for efficient general distribution of cowpea diversity to interested researchers.

G.2.3 Virus cleanup of cowpea germplasm

N.Q. Ng and H. Rossel

Some cowpea viruses are seed-borne such as cowpea aphid-borne mosaic, cucumber mosaic and cowpea yellow mosaic, which are common on-site at IITA. Although attempts were made to rogue out virus-infected plants during field multiplication, it appears that seed stocks of many germplasm accessions still contain virus. For international distribution, it is important that they be cleaned. Accessions suspected to contain virus were planted out in an insect-proof screen house in 1991 and plants exhibiting symptoms were rogued; seeds were harvested from healthy plants only. A complete 'clean-up' of all the existing cowpea collection will take many years because of the size of the collection.

G.2.4 Wild *Vigna* germplasm conservation

S. Padulosi

New *Vigna* accessions are being multiplied and classified and their passport data documented in a computer data base. About 500 new *Vigna* accessions were multiplied in 1991, raising the total to about 1620 accessions from 50 species. The work is ongoing.

G.2.5 *In-vitro* yam germplasm conservation

S.Y.C. Ng, N.Q. Ng and R. Asiedu

Maintenance of the existing 1,500 yam accessions *in vitro* is a major task. In addition a total of over 1,000 new accessions were transferred from the field to *in vitro* meristem culture in 1991. Out of these, over 600 accessions developed into plantlets. Maintenance of the existing collections will be continued and more germplasm will be transferred from the field to *in vitro* culture annually. The effects of different sucrose concentrations in combination with mannitol in the culture media on nodal growth and development were studied on two yam clones. Preliminary results suggest that percent shoot formation was higher in treatments supplemented with 1 to 5% sucrose with or without mannitol, but higher levels of sucrose also inhibited leaf formation and growth. These effects will be further studied in 1992. The effects of mineral oil overlay on yam plantlet growth and development will also be studied.

G.2.6 *In situ* yam germplasm conservation

N.Q. Ng, D. Ogunsola and S.Y.C. Ng

Yam germplasm conservation is an arduous task. Most germplasm accessions do not produce flowers or seeds, thus they have to be maintained vegetatively in field nurseries and/or by *in vitro* culture. Field rejuvenation/multiplication is slow because only a few tubers are produced per plant and the crop growth cycle is long (6 to 9 months). Furthermore, tubers are bulky, requiring lots of space to store; and they lose viability in less than a year, so the whole propagation process has to be repeated annually. In 1991 we found that storing "seed yams" (small tubers derived from "minisets") rather than the large, bulky "ware yams" is much easier and gives a much higher rate of multiplication. Thus during 1991, all existing as well as new yam accessions (over 2,100 from 8 species) were propagated by the miniset technique, on ridges covered with plastic mulch to inhibit foliar diseases and suppress weeds as well as eliminate staking labor. Additionally, shoot tips from over 1,000 accessions that were not previously maintained *in vitro* were sampled in early growth stages for maintenance in the Tissue Culture Laboratory of TRIP. Characterization activity also took place during 1991: about 200 accessions were planted using normal management practices for ware yam production so that data would be relevant to field performance. All these activities will continue in 1992.

G.2.7 Cassava germplasm conservation

N.Q. Ng, T. Nandang, S.Y.C. Ng, R. Asiedu and S.K. Hahn

Cassava cuttings from the TRIP collection were received by GRU in 1991 and planted in a new field at close spacing. It is intended that the materials be re-planted every 9 months or less, to reduce risk of loss. A total of about 1,700 clones were established, including 350 local cultivars from Nigeria, Kenya, Congo and Brazil; 52 clones of wild *Manihot* (24 species); 1275 clones of hybrids between IITA improved varieties and selected clones; and 27 breeding lines from CIAT. Passport data was traced through examination of old field notebooks and stored in a computerized data base. About 400 accessions of existing local cultivars were planted on ridges at normal spacing for characterization for up to 53 descriptors. These activities will continue in 1992.

G.2.8 Rice germplasm conservation

N.Q. Ng, E.S.O. Erimah and J. Obarinde

Although the mandate for rice improvement has been transferred to WARDA, IITA retains responsibility for the germplasm collection for the time being. The rice collection consists of 9,473 accessions of Asian cultivated rice (*O. sativa*), 2,503 of African cultivated rice (*Oryza glaberrima*) and 379 of wild *Oryza* species. About 4,000 accessions have been processed for long-term storage (-20°C, 5% seed moisture content). About 90% of the collection has been characterized with data stored in a computer data bank. During 1991, about 2,500 accessions were multiplied/rejuvenated.

G.2.9 Miscellaneous germplasm conservation

N.Q. Ng, J. Obarinde, A.E. Adegbite and J. Apeji

We maintain collections of 2,000 Bambara groundnut (*Vigna subterranea*), 1,400 soybean, 1,000 maize, 127 African yam bean (*Sphenostylis stenocarpa*), 100 green gram (*Vigna radiata*), 32 Lablab bean (*Lalab purpureus*), 27 winged bean (*Psophocarpus tetragonolobus*) and several pigeon pea (*Cajanus cajan*), jack bean (*Canavalia ensiformis*), and sword bean (*Canavalia gladiata*) accessions; and more than 20 species of multipurpose trees. During 1991, around 50 accessions of miscellaneous legumes including soybean were multiplied/rejuvenated. This will continue in 1992. Thirty-nine clones of banana and plantain collected from Nigeria and maintained as a living collection in a hydromorphic field at IITA-Ibadan in 1991 will be consolidated into the Onne Station collection maintained by PBIP in 1992.

G.2.10 Germplasm distribution

N.Q. Ng and J. Obarinde

We responded to about 150 requests for germplasm or information during 1991. We distributed over 4,500 samples of germplasm to about 20 countries. 68% of the materials were requested by IITA researchers. This is an ongoing activity that will continue in 1992.

G.3 Phylogeny of germplasm

Project rationale

Germplasm is only useful for crop improvement if its phylogenetic position is understood. In recent years, collections from many poorly-defined species have been obtained, particularly from wild *Vigna* and *Dioscorea*. Work is underway to better understand the phylogenetic relationships among our accessions.

Completed studies

Ng, N.Q. and F.M.O. Agbo (unpubl.). Survey of yam diversity in Nigeria.

Through literature survey, field exploration and study of herbarium specimens and plants collected from Nigeria, detailed information on the distribution, ecology and characteristics of all species described or collected were gathered. The information is being prepared for publication to guide future exploration and research work on yam species found in Nigeria.

Ng, N.Q. (unpubl.). Crossability within *Vigna unguiculata*.

Reciprocal crosses were made among 12 genotypes representing all the recognized subspecies of cultivated and wild *V. unguiculata*. Rate of pod setting and pollen fertility in parents and their F1 hybrids were studied to delineate genetic affinity.

Agwaranze, F.N., N.Q. Ng and T.A.O. Ladelinde (unpubl.). Morphological variability within *Vigna vexillata*.

Sixty-six accessions of *V. vexillata* representing 3 botanical varieties have been evaluated for 39 morphological characters. Principal component and cluster analyses were used to determine inter-relationships.

Padulosi, S. and N.Q. Ng (unpubl.). Ecogeographical analysis of wild *Vigna unguiculata*.

About 70 morphophysiological traits measured on approximately 400 accessions of wild cowpea in 1989/90 have been analysed to assess the variability among the taxa in relation to geographical origin. In addition data on localities of about 2,000 herbarium records assembled from herbaria survey in Europe and Africa were also used to produce detailed maps of distribution of different varieties. The studies have thrown light on the centre of diversity of the species and will help in planning future exploration missions.

Activities

G.3.1 Systematics of African *Vigna*

N.Q. Ng

Systematic crossing among different African *Vigna* species began in 1988 to 1) determine genetic affinity between species and clarify taxonomy, 2) determine the gene pool boundary of cowpea and 3) find "bridging species" to help transfer traits from *V. vexillata* to cowpea. Fertilization and embryo development following wide crosses are being tracked as well as cytological and morphological features of the hybrids. During 1991, the *V. oblongifolia* x *V. ambacensis* cross was achieved for the first time ever, as was *V. ambacensis* x *V. luteola*. More crosses are being made and the hybrids are being studied in 1992. Successful crosses between *V. oblongifolia* and *V. luteola* have also been reconfirmed.

G.3.2 Systematics within *V. unguiculata*

S. Padulosi, G. Venora and N.Q. Ng

A cytogenetic analysis of a diallel cross involving all recognized varieties of wild and cultivated *V. unguiculata* is underway to elucidate systematic relationships.

G.3.3 Stomatal variation in *Vigna unguiculata* (§)

S. Padulosi and G. Venora

Characterization of leaf stomata of wild *V. unguiculata* with the use of a computerized image analyser will be carried out in collaboration with the Experimental Station for Cereals, Caltagirone in Italy. The results will determine whether this character is useful for classification purposes.

G.3.4 Pollen/seed anatomy of African wild *Vigna*

S. Padulosi and De Leonardis

In collaboration with the University of Catania, Italy, scanning electron microscope studies on pollen and seed anatomy of African wild *Vigna* is being carried out to clarify taxonomic relationships.

G.3.5 Monograph of African wild *Vigna*

S. Padulosi and N.Q. Ng

Descriptive characterization of African wild *Vigna* will be published as a monograph including information on the distribution, ecology and potential utilization of the various species. Drawings will be published to help identify species in the field.

G.3.6 Identification of yam accessions

R. Asiedu, N.Q. Ng, I. Ekanayake, M. Bokanga and NRCRI

See T.4.2.

G.4 NARS collaboration and training

Project rationale

Plant explorations are always done collaboratively with the NARS of the country being explored. Networks sustain collaboration in a cost-effective manner. Training in genetic resource conservation, evaluation and distribution is essential for the longer-term preservation of biodiversity in Africa.

Activities

G.4.1 Cassava germplasm network (§)

N.Q. Ng and R. Asiedu

A cassava germplasm workshop jointly sponsored by IBPGR, CIAT and IITA will be held in Colombia in August-September, 1992. The workshop will set priorities and responsibilities for collection, conservation and utilization of cassava genetic resources. A NARS representative from East Africa and one from West/Central Africa were recommended to attend this workshop by the participants to the meeting of collaborators in the Root and Tuber Improvement and Production System Research workshop held at IITA in June 1991.

G.4.2 Training course in genetic resources (§)

N.Q. Ng and J. L. Gulley

IITA, FAO and IBPGR will organize a three-week training course in plant genetic resources collection, utilization and management for mid- and upper-level agricultural researchers and technical officers in Africa from September 21 to October 9, 1992. The course aims to strengthen skills in crop genetic resources conservation and utilization.

G.4.3 Graduate training

1. Genetic introgression of *M. glaziovii* into cassava and taxonomic position of tree cassava. N.W. Wanyera (Ph.D. thesis) and S.K. Hahn.

Biotechnology Research Unit

In view of the rapid advances being achieved worldwide in molecular biology and related tissue-culture techniques for gene transfer, the Board of Trustees in 1987 approved the establishment of a Biotechnology Unit at IITA. Its primary role is to identify, introduce and adapt new techniques that can strengthen ongoing research activities elsewhere at IITA, in particular helping to overcome problems that are intractable by conventional breeding and refinements of virus diagnostic methods. Once a new technique is verified and made routine, its routine application becomes the responsibility of the relevant program. Scientists from those programs use the Biotechnology Research Unit's labs to conduct the routine application activities, while the Biotechnology Research Unit staff coordinate lab use and maintenance. The Unit presently houses labs for DNA analyses, virus diagnostics, electrophoresis, cytogenetics and tissue culture.

Another important role of the Unit is to strengthen NARS biotechnology research capacity. Graduate training, training courses, workshops, visiting scientistships, internships and other mechanisms are being adopted to make this happen. The Unit thus serves as a facilitating channel to help the flow of the new techniques and skills to African researchers.

The Head of the Unit, Dr. G. Thottappilly is on sabbatical leave from February 1992 to February 1993. He will spend 6 months at the Scottish Crop Research Institute, Invergowrie, Scotland, studying methods such as PCR cloning, sequencing techniques, *Agrobacterium* - mediated transformation and tissue culture technology to introduce virus resistance genes into crops. His second 6 months will be spent at the Department of Botany and Microbiology, Auburn University (USA), on cDNA cloning and characterization of Tepary lectin genes.

Virus diagnostics activities of the Unit are not listed here; see the Plant Health Management Division workplan. Also note that Plantain biotechnology work, centered at Onne Station, is described in the Plantain and Banana Improvement section (P.3).

B.1 Molecular marker techniques

Project rationale

Molecular marker techniques such as restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), DNA fingerprinting, and electrophoresis are being explored as tools to:

1. Delineate phylogenetic relationships based on DNA similarity;
2. Map genes of economic interest, so nearby markers can be identified and used for more efficient gene transfer;
3. Verify interspecific hybrids; and
4. Give "fingerprints" for the identification of varieties in impact studies.

Completed studies

Terauchi, R., V.A. Chikaleke, G. Thottappilly and S. K. Hahn. 1992. Origin and phylogeny of Guinea yams as revealed by RFLP analysis of chloroplast DNA and nuclear ribosomal DNA. Theor. Appl. Genet. 83: 743-751.

Phylogeny of Guinea yams (*Dioscorea rotundata* and *D. cayenensis*) was studied using RFLP analysis of chloroplast DNA and nuclear ribosomal DNA. This technique enabled us to make a substantial revision in yam phylogeny. Contrary to the general belief that white yam (*D. rotundata*) is a sub-species of yellow yam (*D. cayenensis*), our results indicate the opposite.

Schnapp, S.R., P.M. Hasegawa and G.O. Myers (unpubl.). DNA fingerprinting to detect Interspecific hybrids in *Vigna*.

The *V. oblongifolia* x *V. luteola* hybrid has been verified using the restriction enzyme BamHI and a probe for nuclear-encoded rDNA. However, this enzyme/probe combination did not provide useful polymorphisms when applied to crosses between closer relatives such as cultivated cowpea x wild subspecies of *V. unguiculata*.

Activities

B.1.1 RFLP mapping of cowpea

N. Young (Univ. of Minnesota) and G.O. Myers

An RFLP mapping project is currently underway with the University of Minnesota and should be completed this year. New populations will be developed to help develop greater saturation of the map. F2 populations will be screened at IITA to provide necessary data for identifying linkages of DNA markers to useful genes.

B.1.2 Construction of a yam genomic library (§)

H. Mignouna, R. Asiedu and N.Q. Ng

Chloroplast DNA (cpDNA) RFLP has already been used to study phylogeny of the guinea yam (see "Completed Studies"). We propose to construct a yam genomic clone library and screen these clones to determine which detect polymorphisms in *Dioscorea*. This genomic library could then be used in developing an RFLP map of yam. The cytoplasmic genomes are much less complex than the nuclear genome, and are reliable indicators of genetic evolution. We propose to use conventional electrophoresis and mitochondrial DNA RFLP analysis on the same species.

B.1.3 Molecular phylogenetic studies in *Vigna*

S. Schnapp and H. Mignouna

Phylogenetic studies in *Vigna* are being pursued using DNA markers in an effort to facilitate the design of wide crossing schemes for improvement of cowpea. A crossing study was also conducted to assess the reproductive affinity between *V. oblongifolia*, *V. luteola*, and *V. unguiculata*, using three accessions from diverse geographic locations for each species.

B.1.4 RFLP detection of hybrids in *Vigna*

S.R. Schnapp, G.O. Myers and N.Q. Ng

There is an ongoing effort to identify useful enzyme-probe combinations that will reveal polymorphisms in genomic DNA of *Vigna* species. These are being used to verify putative interspecific hybrids and to clarify phylogenetic relationships.

B.1.5 RAPD analysis to detect *Vigna* hybrids (§)

S.R. Schnapp, G.O. Myers and N.Q. Ng

Previous work (see "Completed Studies") found that RFLP's did not detect differences between closely related subspecies of *V. unguiculata*. A more sensitive method known as Random Amplified Polymorphic DNA (RAPD) has recently become available and will be tried. In RAPD analysis, a DNA polymerase enzyme (Taq polymerase) specifically amplifies certain DNA sequences (using the polymerase chain reaction, or PCR) corresponding to the DNA primer sequence provided during the reaction. Since small DNA sequences can be used as primers (e.g. 10 base pairs), relatively small differences in DNA can be detected.

B.1.6 RFLP to detect germplasm duplicates (§)

H. Mignouna and N.Q. Ng

The IITA germplasm resource unit has about 15,000 accessions of cowpea and related wild species. In order to eliminate duplications, these accessions need to be characterized and classified. To determine the feasibility of using RFLP to detect duplications, a pilot investigation on 100 selected accessions will begin in 1992.

B.2 Non-sexual gene transfer techniques

Project rationale

Our conventional cowpea, yam and plantain improvement work now faces substantial difficulties in sexual gene transfer due to shy or asynchronous flowering, genome incompatibilities, hybrid embryo dysfunction and hybrid sterility. Somatic cell callus and protoplast culture followed by transformation or somatic hybridization are techniques for bypassing the sexual process. These techniques could thus make a substantial contribution to improving IITA's crops. Basic work, involving close collaboration with advanced laboratories elsewhere, is underway to establish these techniques.

Completed studies

Ngu, M.A., T.A.D. Ladelinde and S.Y.C. Ng (unpubl.). Embryo culture and callus induction in yams. Mature embryos of two yam species, *Dioscorea rotundata* and *D. abyssinica* were cultured on six different culture media: Linesmaier and Skoog (LS) medium, MS medium, Nitsch and Nitsch medium, White medium, 1/2 strength LS and 1/2 strength MS solid medium. Nitsch and Nitsch medium was superior to the other five media for embryo germination. Embryo germination was very poor in White medium. The germinated plantlets were successfully transplanted and established in soil. There were different responses to the media between the two species. Embryos of *D. rotundata* and *D. abyssinica* were also used as explant for callus induction. The excised embryos were cultured on MS medium supplemented with different levels of 2,4-D. Callus formation was obtained in medium containing 9 μ M, 18 μ M and 36 μ M 2,4-D. Root formation was obtained after transferring of the callus to 2,4-D free medium. Sporadic shoot formation was obtained in *D. rotundata*.

Activities

B.2.1 Somatic embryogenesis in cassava

S.Y.C. Ng

Media for somatic embryogenesis from callus derived from young cassava leaves has been developed. Over thirty IITA improved cassava clones were screened in 1991 for their ability to produce somatic embryos *in vitro*. MS medium supplemented with different levels of 2,4-dichlorophenoxyacetic acid (2,4-D) were used. Globular embryos were obtained from six clones after 20 days in culture. The embryos were then transferred to Murashige and Skoog (MS) medium with different levels of zeatin for maturation. Embryos with green cotyledons were obtained from all the six clones. However plant regeneration was obtained from only four clones. Efforts will be made in 1992 to increase the number of genotypes that can be regenerated and the plantlet regeneration rate per genotype.

B.2.2 Callus induction and regeneration in yam (§)

S.Y.C. Ng

A callus culture and plant regeneration system for yam is a prerequisite to achieving *in vitro* gene transfer. Different types of tissue explants and a range of genotypes of *D. rotundata* (and later, of *D. alata*) will be cultured in media to induce direct somatic embryogenesis or callus formation. Callus will be subcultured and plant regeneration attempted.

B.2.3 Callus induction and regeneration in cowpea

S.Y.C. Ng, G. Thottappilly and N.Q. Ng

In many cases it is observed that genotype has great influence on the response of explants to *in vitro* culture. Initially, 10 selected genotypes of cowpea will be used. Young cotyledons and epicotyls will be cultured in media containing different levels of 2,4-D, NAA with or without kinetin. Subsequently, the callus will be transferred to regeneration media.

B.2.4 Anther culture in cassava

S.Y.C. Ng and K.V. Bai

Anther/microspore culture can be used to produce haploids and di-haploids, which would be very useful for genetic studies in cassava. Anthers of more than 10 cassava clones and two related wild species were cultured on MS medium supplemented with picloram or 2,4-D. All clones produced callus in all treatments. However, green protocorm structures and roots were obtained from only three clones, cultured on MS medium supplemented with 2,4-D. Further studies will be conducted on more genotypes and microspores at different stages of development.

B.2.5 Resistance bioassay for pod-sucking bugs

L.E.N. Jackai, R.E. Shade (Purdue Univ.) and S.R. Schnapp

Since regeneration of cowpea plants from callus remains problematic, a callus bioassay system has been developed for *Maruca testulalis* and efforts to develop the system for application to pod-sucking bugs are underway.

B.2.6 Protoplast fusion in cowpea

E. Fillipone (Univ. of Napoli)

Protoplast isolation and multiplication techniques have been developed and somatic fusions have been made. Tolerance of cowpea to abiotic stresses is being studied on protoplasts and free cells.

B.2.7 *Agrobacterium* gene transfer in cowpea

S.Y.C. Ng, G. Thottappilly and N.Q. Ng

Genetic transformation using *A. tumefaciens* carrying the Kanamycin resistance gene will be attempted using leaf tissues, cotyledons, epicotyls, embryos and meristems of cowpea.

B.2.8 DNA particle gun transformation in cowpea

R.A. Bressaw, P.M. Hasegawa (Purdue Univ.),

S.R. Schnapp and P. Dunn

Transient gene expression has been observed in cowpea transformed through DNA microprojectile bombardment. While more refined protocols are being developed, the construction of vectors, initially with Bt toxin genes is underway.

B.3 New cytogenetic techniques

Project rationale

Manipulation of polyploids has become important in cassava, yam, plantain and cowpea breeding at IITA (see relevant chapters). While conventional cytogenetic methods have been used for decades, and continue to have critical applications at IITA, new methods are also arising which can extend our capabilities in this classical area of plant breeding.

Completed studies

Hahn, S.K. (unpubl.). Standardization of DNA flow cytometry to determine ploidy in cassava.

Recalcitrant flowering is not unusual in cassava, impairing ploidy analysis of reproductive cells. DNA flow cytometry analyzes vegetative tissue, overcoming this bottleneck. Leaves at different growth stages were harvested from cassava plants of different ploidy levels (2x, 3x, 4x) for analysis. Recently fully-expanded leaves gave better results than younger or older leaves. The technique enabled separation of an aneuploid of $2n = 36+8$ from the rest of the polyploid groups. Different genomes may have different masses, so in theory autopolyploids can be distinguished from allopolyploids. We were able to verify this. Somatic autotetraploids showed two distinct peaks while sexual polyploids usually showed just one major peak. Thus, as well as identifying the number of complete genomes present (ploidy level *per se*) and aneuploidy, the method seems capable of distinguishing important classes of polyploids from one another.

Activities

B.3.1 DNA flow cytometry in cassava

S.K. Hahn, K.V. Bai and S.Y.C. Ng

Polyploid cassava clones have been produced from diploid interspecific crosses with wild *Manihot* species. DNA flow cytometry is being used to facilitate ploidy determination. Aneuploids are being produced for genetic studies.

B.3.2 DNA flow cytometry in yam

S.K. Hahn, K.V. Bai and R. Ugborogho

Polyploidy is being delineated in yam by testing open-pollinated seed of different ploidies using DNA flow cytometry. All breeding accessions including wild species are being screened for ploidy; up until now, it has been found that yams vary from 2x to 16x. Crosses are being made to understand the potential for introgression and details of phylogeny.

B.4 NARS collaboration and training

Project rationale

One role of the Biotechnology Research Unit is to act as a bridge for the flow of useful biotechnology techniques to African NARS.

B.3.2 Graduate training

1. Phylogeny of wild and cultivated *Manihot* species based on restriction fragment length polymorphism. M.A. Fregene (Ph.D. thesis) and R. Asiedu.
2. *In vitro* culture of anthers and pollen grains of cowpea. U. U. Ekuere (Ph.D. thesis) and S. R. Schnapp.
3. Isolation of rice yellow mottle virus: characterization of genomic and subgenomic RNA. D. Beimo (B.Sc. thesis) and G. Thottappilly.

B.3.2 Workshop

1. Plant Cell and Tissue Culture and Biotechnology Workshop. Ibadan, April 13-24, 1992.