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Progress Report to January 1970

INHERITANCE AND IMPROVEMENT OF PROTEIN QUALITY AND CONTENT IN *SORGHUM VULGARE* PERS.

Prepared by

R. C. Pickett
Department of Agronomy
Purdue University

Contract AID/csd-1175

U. S. Agency for International Development,
Department of State
with the
Purdue Research Foundation,
Lafayette, Indiana

SORGHUM PROJECT STAFF - 1969

R. C. Pickett, Principal Investigator
Professor, Agronomy

E. T. Mertz
Professor, Biochemistry

D. T. Cooper, Post-Doctorate
Agronomy

R. Jambunathan, Post-Doctorate
Biochemistry

Fred Collins
Graduate Research Assistant (3/4 time this project)
Succeeded by D. P. Mohan, (India) September 1, 1969

Greg Hinze
Graduate Research Assistant (sorghum testing 3/4 time)
succeeded by Dallas Oswalt, September 1, 1969

A. O. Abifarin
Graduate student (Kansas State-Nigeria-USAID Contract)
terminated June 1, 1969

Fernando Rodriguez
Sabbatical leave from the University of Puerto Rico
terminated June 1, 1969

Samiri Mukuru
Graduate student - Rockefeller Fellow - Serere, Uganda
began September 1, 1969

Bantayehu Gelaw
Graduate student - Rockefeller Fellow - Alemaya, Ethiopia

Rameshwar Singh
Graduate student - Rockefeller Fellow - U.P.A.U. India
began September 1, 1969

Robert Schaffert
Graduate student

R. C. Worley
Field Technician

Shirley Tyner
Laboratory Technician

Kathleen Flood
Assistant Laboratory Technician

Ardith Spies
Administrative Assistant

"INHERITANCE AND IMPROVEMENT OF PROTEIN QUALITY AND CONTENT IN *SORGHUM VULGARE* PERS."

Introduction

Sorghum is an extremely important human food crop in much of tropical Africa and parts of Asia. Throughout the underdeveloped world it has potential perhaps as the leading feed grain for animals since the other major cereal crops including rice, wheat and corn are used almost entirely for human food. Recent research at Purdue University and elsewhere has shown that extremely high yields of grain are possible. Farm production has achieved between 10,000 to 12,000 lbs/A while experimental plot yields have been between 15,000 and 20,000 lbs/A. These are under extremely favorable conditions, but perhaps yields of 2,000 to 5,000 lbs/A under moisture stress may be an even more important contribution in areas of developing countries where they are most needed. These yields are all compared to unimproved agriculture yields of 500 to 700 lbs/A.

Simultaneously with these yield increases, work during the last three years on this project has shown that improved protein levels and amino acid composition improvement are quite practical. In contrast with the normal protein levels of 7 to 9 percent protein it appears that 13 to 15 percent is obtainable in many relatively high yield varieties and hybrids. Amino acid composition may be simultaneously improved with the limiting amino acids increased 50 to 100 percent. Protein levels of 15 to 26 percent have been found in lower yielding less adapted types, but these show that even higher levels may be obtained in high yielding form in the future. Studies in 1969 gave the most assessment of yield and quality combination in present commercial sorghum hybrids while studies coming in 1970 will give a comparison of what is possible with new generation materials.

Since research on food and feed crops is very limited on crops other than rice, wheat and corn a great deal of energy has been expended to get further research programs developed or extended in each interested country in South America, Africa and Asia. Sorghum research in the tropics received a great boost when the world sorghum collection was first assembled by The Rockefeller Foundation and many cooperators between 1958 and 1963. Additons to this collection have been made by this project and many cooperators since then. Research programs have been greatly expanded especially in India and also in Thailand, Uganda and Nigeria. USAID and Rockefeller assistance has played a key role in many of these research efforts. The contribution from this program to these and many other increased research efforts encompasses the sending of new experimental seed, information on chemical analysis and breeding behavior and research planning and advice mostly through visitation by the principal investigator of this project. In return seed from other locations has been submitted for chemical analysis at Purdue and this activity will greatly increase in the future. In addition, information on performance of these new materials is requested to be sent back to Purdue for analysis.

The new research efforts by this project at this point center around further clarification of the mode of inheritance of protein and level of amino acid composition and finding out the interactions with various environments. Information to date makes it quite certain that many genes influence protein level and that amino acid composition may be shifted by several major genes in the endosperm with additional modifications being possible through shifts in proportion embryo and its composition. The embryos average approximately twice the protein amount and up to three times as much limiting amino acid concentration as the endosperm. Additional factors that deserve considerable attention, in addition to yield and chemical composition, include

digestibility, where the range seems to be as much as 40 to 80 percent digestion by experimental animals, weathering resistance and storability. Local selection is under way for acceptability and palatability for the use intended as well as resistance to the local environmental conditions, diseases and pests.

Research Objectives

This project on sorghum improvement had as its initial technical research objectives the following:

a) To screen for high lysine and nitrogen level in selected lines from the world sorghum collection and various breeding programs in the U.S., Africa and India. Crosses and inbreds from them would also be examined to determine the inheritance pattern.

b) To examine threonine, tryptophan, methionine and isoleucine (high ratio with leucine) contents and determine the complete amino acid distribution on select high lysine lines and recombined lines and hybrids among them. The inheritance of the levels of each will be analyzed.

c) To recombine the lines with highest levels of essential limiting amino acids with highest level protein contents possible in high yielding inbred lines and hybrids.

d) To determine biological or nutritional value of high protein, high quality amino acid selections.

e) To study the chemical nature of proteins on sorghum endosperm and embryo. Sorghum selections with the greatest differences in amino acid composition would also be studied.

The major way in which the results of the project could affect overseas agricultural improvement was conceived as a system of contacting interested research workers and to develop interest in sorghum improvement in all countries where sorghum would have use as a food or feed grain (i.e.South America, Africa and Asia). Improved seed, information including chemical analyses and research planning advice is sent to all interested countries.

Purdue Sorghum Project Research Reported To Date

Since 1963 repeatedly in various types of trials, yields of 15,000 to 20,000 lbs/A have been noted. Almost all of these yields that are significantly above the yields of 10,000 to 12,000 lbs/A occur in hybrids in the six to nine foot height class and have significantly larger heads. Studies continue on taller, larger headed materials. The initial work on quality in 1966 (report no. 1) was done on diverse ranges of germplasm regardless of economic degree of adaptation. As a result the range of protein from 7 to 26 percent and lysine from .7 to 2.8 percent was immediately uncovered and little further variation in protein amount or lysine has been seen since then. The exception being that lysine was subsequently found to be under .5 and up to 3.8 percent.

Work by Campbell (report no. 2) showed that soil nitrogen level could significantly influence the amount of protein but that the genetic effect was still much greater than the adjustment due to fertilization. This study and one by Collins indicated that there was a genotype x location interaction for protein and lysine. This means that while selection can surely be effective for higher protein, it will be necessary to test extensively under various environments to determine the benefits for any given location.

Heterosis for grain yield has been described by Collins and Abifarin and others as occuring up to 150 percent with levels over 50 percent occuring with high frequency. Heterosis for protein percent can occur but in the crosses tested to date it occurs rarely. The most frequent phenomenon is that the level of protein in the parents is transmitted to the hybrids at the same or slightly lower level.

Collins studies reported in 1968 (report no. 4) indicated that there were several genes that influence protein level and lysine levels and that both additive and non-additive genes affected both protein and lysine as well as yield and that protein and lysine can be incorporated into high yields. Lysine levels also transmit well into the hybrids, but rarely is the hybrid higher than either parent and usually follows the same level as the parent. It would seem to be efficient to select for high lysine or protein in the variety itself and then use it either as an open-pollinated variety or as a parent in hybrids where they may be produced.

Digestibility is another important factor with a large range of variability and much possibility for improvement. Beginning in 1966 studies verified this. This was initially seen in a range of coefficients of digestibility in rat feeding trials from 49 to 77 percent. Subsequent rat feeding trials showed a range of protein efficiency ratio of .18 to .91 percent. In 1969 <u>in vitro</u> digestibility ranged from 50 percent dry matter digested to 77 percent on a limited range of commercial hybrid germplasm (report no. 6). This compares favorably with the range of 30 to 73 percent digestibility reported by Anthony and co-workers at Auburn University which was done on a wider range of germplasm. Work by Isis Nawar (report no. 2) showed a range in digestibility from 49 to 77 percent and relative nutritional value

expressed as a percent of 21 to 39 percent.

Studies on the type of sample needed for chemical analysis of grain showed that a mixture of several open-pollinated heads in the line or hybrid was the best. There is a considerable variance from plant to plant and treatment factors such as flag leaf, pollinating bags or other modifications can seriously affect seed development and yield as well as chemical composition.

Data released in report no. 4 for work through 1968 reported for several hundred lines in 1968 and showed the variation in the other limiting amino acids to be considerable.

2.0 to 5.5 for threonine

.34 to 4.51 for tryptophan (the lowest levels may involve destruction, methodology still in development stage)

.98 to 4.62 for methionine/cystine (sulfur-bearing amino acids)

3.26 to 5.51 for isoleucine

9.5 to 17.1 for leucine

.272 to .380 for isoleucine/leucine ratio (highest ratio desirable)

There is some evidence that high levels of leucine are deleterious per se. A catalog of protein (N) and lysine values of collection accessions is being prepared by this project. Complete amino acid analyses are normally done only on lines that are outstanding in this initial screen for protein (N) and lysine. The levels of three other amino acids of nutritional value have been reported regularly and these are phenylalanine, tyrosine and valine.

Embryo size and composition has been shown to be very important. Studies beginning in 1968 and 1969 (report no. 5) and continuing currently show that the embryo percent in the seed varies greatly. The initial range found was 9.7 to 14.1 percent of the seed as embryo. The protein in the embryo was 18.1 to 24.7 percent and the lysine 5.5 to 6.5 percent of the

protein. Thus on the average the embryo has almost twice the percent protein as the endosperm and almost three times as much lysine.

Other seed characteristics of importance are oil percentage, which is important for energy and as an indicator of protein and seed size with a possible contribution to yield or quality. Mineral composition is being studied as a factor of possible importance. Oil percentage has varied significantly from 1.2 to 5.7 percent and since all of these embryo studies have been done on small amounts of genotypes it is hypothesized that much greater ranges exist in a wider range of material. Seed size is another factor that varies over ten-fold and studies are now underway to determine the interaction of seed size with chemical composition factors.

Studies in 1969 (report no. 6) were done on present U.S. commercial grain forage and sudan hybrids to see the range and combination of yield, protein percent and mineral composition. This work was done on grain, leaves and stems. The grain yields varied from 6,300 to 14,000 lbs/A while the total plant varied from 24,200 lbs/A at the highest to 14,100 lbs/A at the lowest. The protein percent in the grain varied 8 to 13 percent (corn at 8.7 percent) while the protein yield of the grain varied from 615 lbs/A on check RS610 to 995 lbs/A on a high yielding experimental hybrid with 9.5 percent protein and 14,000 pounds grain per acre. In high population plots an even higher protein yield of grain with an intermediate forage hybrid NK300 produced protein yield of 1125 lbs/A, protein percent of 11.2 and grain yield of 11,660 lbs/A.

Purdue Sorghum Project Investigations Currently Underway

A major emphasis in current investigations is being placed on selecting and increasing of high protein and high lysine lines as well as those that are outstanding in other limiting amino acids. Environmental interactions are being intensively studied by means of seed returned from overseas locations for analysis and by tests at four major locations in Indiana. Increase is being made of selected lines for further testing and use by overseas cooperators next year.

Several thesis studies are concerned with the inheritance of protein and amino acid composition in both the embryo and endosperm. Other studies are concerned with embryo size and composition and total seed size which varies over ten-fold.

The factors of height and head size are well known for their association with high yield. These are being studied with relation to use of new diversity of germplasm from the world collection. One thesis is trying to develop selection indices for use in determining which morphological and physiological characteristics are the most useful in selecting new material from the world collection. This is in association with a practical case of a study of wide sampling from the collection.

Another study is concerned with the contribution of tillering. This was found to be more significant in contributing to yield in certain lines than was formerly believed. Previous studies have shown lodging resistance to be quite heritable and governed by a few major genes. These genes will be transferred to the new high yield, high protein lines for practical testing.

Two other factors will be studied more intensively this year. These

include weather and bird resistance, either of which may be critical in sorghum production.

Overseas Cooperation

The present network of cooperators includes workers in 52 countries. The number of nursery sites in each country varies from one to four with a total of 103. Some members of this network were previously known by the principal investigator of this project and some were cooperators in the international sorghum program based in India. The majority, however, were developed as a result of research planning trips by the principal investigator to Africa and Asia in the fall of 1966, 1967 and 1969. South America locations were included in the spring of 1967, June and July of 1968, spring of 1969 and January 1970. Correspondence continues with workers at each location. Four types of contact were developed with each research cooperator.

1) Distribution of selected seed from the world collection and various breeding sources is provided regularly. As each cooperator was contacted and agreed to work on this project of increased yield and nutritional quality of sorghum certain relatively small amounts of seed were given to them from the nursery in 1965 through 1968 depending on when the person was contacted. Preliminary results from these earlier grow-outs have been received and affected the entries planted in 1969 when a major increase was made of all past outstanding performers for yield and various nutritional characteristics. From the 1969 Purdue nursery 12 sets of 400 inbred lines, 47 sets of 300 lines and 44 sets of 200 lines were sent to the various cooperators in

Selections from the World Collection made at Purdue are analyzed for yield and protein amount and quality and then distributed around the world.

accordance with their capabilities and desires. Only inbred lines or non-hybrid open-pollinated materials were sent since this gives the opportunity to select for best adaptive material for open pollinated as well as parents for hybrids where they might be used. Selection was made on the basis of yield potential, combining ability in hybrids, high protein, and improved amino acid profile. This made a total of 103 sets sent to 52 countries. Cooperators included all who expressed interest in countries receiving USAID assistance and workers in strategic "developed" countries. Locations in the following countries received seed:

Argentina	Botswana	Afghanistan
Bolivia	Cameroun	Ceylon
Brazil	Chad	India
Chile	Congo	Indonesia
Colombia	Ethiopia	Japan
Costa Rica	Ghana	Jordan
Dominican Republic	Ivory Coast	Korea
Ecuador	Nigeria	Lebanon
Guatemala	Rhodesia	Malaysia
Guyana	Senegal	East and West Pakistan
Jamaica	Sierre Leone	Philippines
Martinique	Sudan	Taiwan
Mexico	Uganda	Thailand
Nicaragua	United Arab Republic	Turkey
Paraguay	Upper Volta	Viet Nam
Peru	South Africa	
Puerto Rico		Australia
Trinidad		
Uruguay		
Venezuela		

2) Information was given to each worker in separate outlines including "Sorghum Management Suggestions", "Procedures to Follow in Developing Sorghum Breeding Nurseries" and "Information Desired on All Sorghum Distributed from Purdue University". These are available under separate cover. Research reports of initial work completed by this project are distributed directly to cooperators in order to accelerate the information exchange and research programs faster than by usual channels of publication. In the near future these will include information from the other locations. In addition to research cooperators, these progress reports are sent to a current total of 508 research workers in 57 countries, plus a distribution of 125 by USAID. This mailing list is available to interested persons by request.

3) Research planning is accomplished primarily through visits by the principal investigator to the experiment stations with the research personnel concerned. USAID personnel act as consultants and expeditors and in some cases, staff are available to do research on material directly. Follow-up correspondence allows further development of the research plans. Information exchanged from locations other than Purdue is often times an essential function. Information from other countries and even from stations within a country is often not available to the concerned workers.

4) Notes from the nurseries are taken and returned and seed of selected lines is sent to this project for chemical analysis. Many locations have been offered 15 to 20 complete amino acid analyses as an initial step to determine the most valuable sources of protein lines. Analyses have been done on approximately 200 samples and 200 more samples are partially complete. Nitrogen and lysine analysis will be run on 300.

Additional new lines of sorghum are being added to the world collection after being received by this project. These include 2,470 lines from 20 different countries. The present world collection in India already totals almost 12,000 and is increasing. This collection as gathered and disseminated by The Rockefeller Foundation has been included in these studies.

A. Africa

Sorghum is a native of Africa and is the most important human food crop in several countries south of the Sahara Desert. Together with bull-rush millet (Pennisetum typhoides) it makes up the primary food grain from the Sahara down to the humid tropical zone as well as in several other areas including eastern highlands of Ethiopia. There are centers of diversity in both east and west Africa as well as in the south. There are good breeding programs in Bambey, Senegal; Saria, Upper Volta; Kano and Zaria, Nigeria; Serere, Uganda; Alemaya, Ethiopia; Khartoum, Sudan and Guetala, Cameroun. The primary role of this project has been to assist in the selecting and distribution of select high protein, high yielding diverse germplasm in association with The Rockefeller Foundation. The analytical information from this project is on lines selected for high yield potential and also high protein and improved amino acid composition. The severe disease and insect problems necessitate selection on these in the Africa improvement program, but it is hoped that through distribution of germplasm and aid in analytical work that the next improved germplasm can be improved in nutritional quality as well as yield. Many other locations do not have major research programs, so encouragement and planning by this project has assisted in getting several programs organized into greater activities. These include stations in Sierre Leone, Ivory Coast, Ghana, Chad, Congo and the UAR. Mr. Sam Mukuru from

Sorghum at Alemaya, Ethiopia. Mr. Bantayehu is working on quality investigations at Purdue in preparation for further improvement work in Ethiopia.

Serere, Uganda and Mr. Bantayehu from Alemaya, Ethiopia are presently working on this project as graduate students on thesis problems dealing with protein quality and yield. Dr. Melak Mengesha, now Dean of Agriculture at Haille Selassie I University at Alemaya, Ethiopia, received his Ph.D at Purdue with this project and is actively cooperating with this improvement work and acquisition of additional germplasm. Dr. Ayo Abifarin, now with the International Institute of Tropical Agriculture at Ibadan, Nigeria, also received his Ph.D at Purdue with this project.

Dr. Abifarin (Purdue Ph.D 1969) in sorghum nursery at Kano, Nigeria where yield and quality increases are being sought.

N.G.P. Rao, Coordinator of the India sorghum program. Amino acid analyses have been made at Purdue by this project for their program.

B. Asia

In India a world center of activity is organized with cooperation of the Indian government, several state agricultural universities and The Rockefeller Foundation. Work by this project is completely cooperative with the work of this center through participation in joint international nursery distribution from India and analyzing for nitrogen and lysine as well as complete amino acids for several experiments in India. Thereby there is an acceleration of the interest and development of improved nutritional quality with the next improvement of yield, new varieties, and hybrids in that program. The world collection of sorghum assembled there has been distributed to cooperators around the world including this project. The Rockefeller Foundation coordinator, Dr. Lee House, spent his study leave in 1968-69 here at Purdue University cooperating with this project. Two graduate students from India, D. P. Mohan and Rameshwar Singh, are presently working with this project on their Ph.D studies.

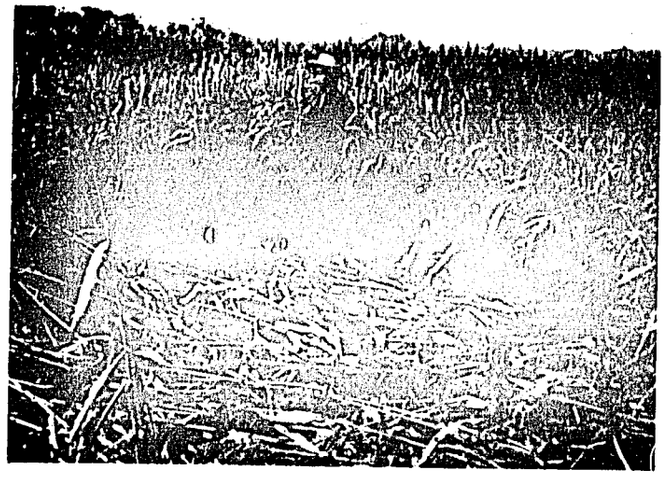
In Pakistan sorghum has been an important human food crop for several million people but research has been very limited. A meeting of the principal investigator of this project with their technical leader and the Minister of Agriculture resulted in additional germplasm being sent from

this project and selection started for improvement in both nutritional quality and yield as well as resistance to diseases and pests.

Additional grain production is needed in Afghanistan on many irrigated sites but there has been no effective sorghum improvement work previously. Currently an attempt is being made to organize new research and planning with USAID personnel and local staff with new germplasm being grown out each year. The hope is to follow the pattern of new production practices similar to the new rice and wheat work.

A major emphasis in the total agricultural improvement program in Turkey centers around feed grains for which sorghum is the primary nominee. A major thrust by the USAID personnel and the Ministry of Agriculture is being made to develop new high yield, high quality sorghum adapted for use there.

In Thailand The Rockefeller Foundation in conjunction with the Thailand government has had an excellent program at the Suwon Farm on upland near Bangkok for the last three years. The coordinator of sorghum research for the Kasetsart University is Dr. Banjerd Boonsue who received his Ph.D training with this project. This program has used a tremendous amount of diverse germplasm in very wide crosses and has been an excellent source of segregating material for use at Purdue by this project in finding new high yield, high protein



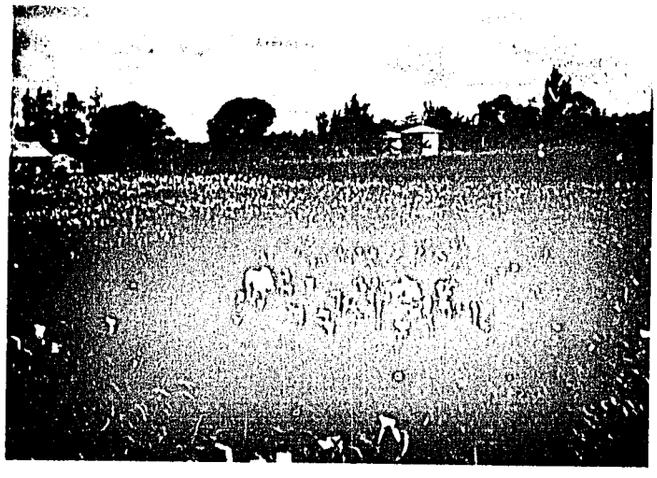
Sorghum at Suwon Farm in Thailand. This program has selected disease and insect resistant lines and furnished diverse segregating seed of much value in other locations. This project is analyzing many lines from there for protein and amino acid composition.

types. The most pressing problem in S.E. Asia seems to be the resistance to diseases and pests. Excellent progress is being made in selecting new resistant material which is being tested in other locations in S.E. Asia. This program is aiding by analyzing for protein and amino acid composition and sending our selections for high protein and yield as additional sources of germplasm. A relatively small percentage has been adapted, but several valuable additions have been made.

Seed exchange and information is being supplied to additional breeding locations in Malaysia, Indonesia, Philippines, Taiwan, Korea, Jordan and Ceylon. In management work of sorghum in the Philippines Dr. Isaac Cagampang is conducting valuable research following his Ph.D training with this project. Great interest is being expressed in expanding the work of each of these locations and the services of this project are being offered to assist in the objective of producing high nutritional quality grain in the next round of improved hybrids.

C. Central and South America

Seed has been sent into each country of South America except Surinam and French Guiana. In addition cooperative work is underway with workers in Jamaica, Dominican Republic, Puerto Rico, Costa Rica, Nicaragua, Mexico



The portion of the world collection that will not produce seed at the Purdue nursery is grown in a winter nursery at Isabela, Puerto Rico.

Sorghum nursery at Santiago, Chile. Material and chemical analyses are being furnished to this program along with many other locations in South America.

and Guatemala. Sorghum is a relatively new crop in all of South America except Argentina where it has been used as a feed grain extensively for 30 years or more. Its distribution is currently spread to the northern part of Argentina into the Chaco area, into southern Brazil and to the west into Chile. In Chile good investigations are underway with both federal and state workers. Perhaps the most rapid progress in yield and quality investigation is taking place with both commercial companies in Argentina and the federal institution called INTA.

Interest is great in the rest of South America, particularly Colombia and Venezuela where the poultry and swine industries are increasing and the feed companies, such as Purina, desperately need efficiently produced

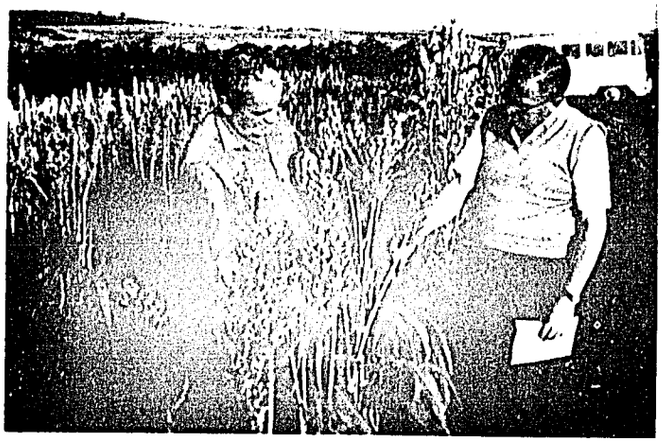
Purdue sorghum selections showing outstanding yield performance in commercial breeding nurseries at Salto, Argentina

feed grain. Relatively new programs are being established in other stress areas.

Experiments in breeding and production of sorghum are being made both east and west of the Andes in Ecuador, Peru and Bolivia. Several new sorghum improvement programs are being established including Paraguay and Uruguay and in the extensive dry northeast zone of Brazil. In addition to the seed that has been sent, research reports have gone to workers concerned, and personal counseling on research plans has taken place.

In Brazil the most rapid progress is being made with Agroceres which sells the majority of the hybrid corn seed in that country. Two years ago they began major investigations in sorghum to provide seed grain for the rapidly developing poultry and swine industries. They sent one of their staff, Paulo Ribas, for 6 months training with this project and have had two nurseries per year with simultaneous efforts at progress in both yield and protein amount and quality. Many of their samples have been analyzed here.

A much higher frequency of adaptation has occurred in the samples distributed to date in Argentina and southern Brazil where relatively high yields are obtained at greater latitudes. The real opportunity for helping to stabilize a whole region may lie in northeast Brazil where new programs are being supported



Sorghum breeding program at Agroceres in Jacaraezinho, Parana, Brazil. Paulo Ribas, who trained for 6 months at Purdue, is now in charge of this program.

by USAID and the Ford Foundation in Recife and Fortaleza. Here heat and drought resistance along with improved yield and quality will show every prospect of making a significant contribution to the entire economy of that area.

In addition to the feed grain contribution in Northeast Brazil, which is the biggest poverty pocket in the Western Hemisphere, sorghum offers a suitable grain that could be eaten directly in case of emergency such as occurs widely in that area. It can be used to mix with wheat to extend and even improve the nutritional value of that relatively expensive grain which still needs to be imported. Presently cassava, with its very low protein content is being used for this supplemental additive.

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No. 1 July 1, 1966 to December 31, 1966

No. 2 January 1, 1967 to June 30, 1967

No. 3 July 1, 1967 to December 31, 1967

No. 4 January 1, 1968 to December 31, 1968

No. 5 January 1, 1969 to June 30, 1969

No. 6 July 1, 1969 to December 31, 1969

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