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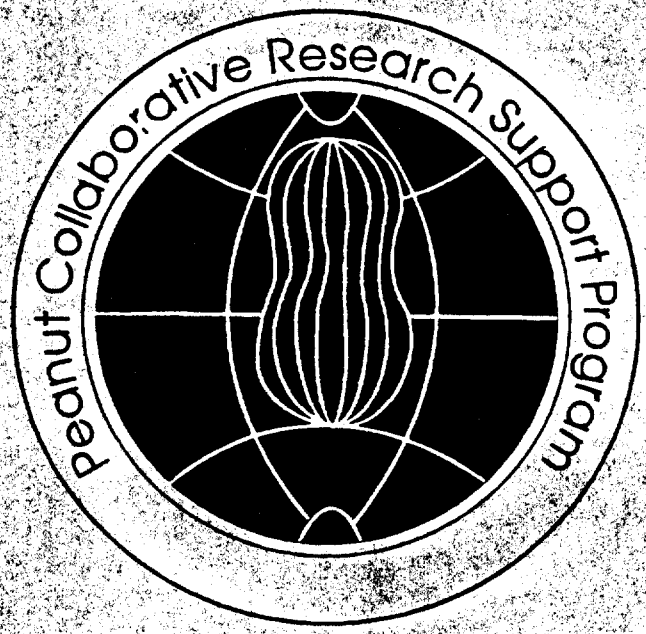
IMPROVING GLOBAL PRODUCTION AND USE OF PEANUT

For Economic Growth, Human Health and Nutrition, and Environmental Sustainability



1995 Annual Report

The Peanut Collaborative Research Support Program

United States Agency for International Development Grant No. DAN-4048-G-00-0041-00

About the Peanut CRSP

Peanut (groundnut) is a crop that enhances sustained, profitable, and environmentally safe use of land with great potential for meeting three food security goals:

- a widely adapted legume crop that increases available food,
- a stable source of cash income for growers, processors, and distributors which increases access to food, and
- provides a better nutrient balance for the user.

The Peanut CRSP (Collaborative Research Support Program) is dedicated to enhancing these potentials through addressing global problems in peanut production and use. Major goals are to generate solutions to current problems and to improve research capabilities that enhance the probability of effectively addressing future problems.

The program involves four U.S. universities engaged in collaborative research with institutions in some 12 developing countries that host the Peanut CRSP, six regional and international organizations, and over 65 scientists, all engaged in priority research and development issues in Semi-Arid Tropical West Africa, Southeast Asia, and the Caribbean. Additionally, the collaborative nature of the program allows for feedback that provides solutions to producer, processor, and consumer problems in the U.S. The Peanut CRSP communicates information on technologies as they are developed through workshops, networks, training and publications.

The Peanut CRSP was implemented in 1982. It is supported by a grant from the United States Agency for International Development to The University of Georgia with sub-grants to the participating institutions. There is a strong and successful effort to insure complementarity with ICRISAT in program planning, technology development, and communication of information.

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EXECUTIVE SUMMARY

INTRODUCTION

The Collaborative Research Support Program (CRSP) was created to implement Title XII of the United States Foreign Assistance Act of 1975. The goal of Title XII is to prevent famine and to establish freedom from hunger through land-grant university involvement in international development. To help attain these goals, the Peanut CRSP was established in 1982 to enhance the research capability of developing countries through training and research support, and to support research at U.S. land-grant institutions.

Peanut is grown in most countries, both developing and industrialized, from 40°N to 40°S latitude. Biological and physical environmental constraints to peanut production and use are similar worldwide, which gives great potential for mutually beneficial collaborative research to relieve these constraints.

As world grain production has reached levels consistently near demand, there has been a shift in consumer demand from staple grains to more nutritive, high-protein foods such as peanut. Improved peanut productivity and processing can improve human nutrition and economic development in semi-arid and sub-humid tropical and subtropical environments. Furthermore, peanut cultivation may enhance the agricultural environment as it plays a significant role in increasing the sustainability of agricultural systems.

BENEFITS OF PEANUT PRODUCTION AND USE

Environmental enhancement

- The peanut crop canopy provides nearly full ground cover. Whether grown alone or in sequence with other crops, peanut cultivation reduces exposure of soil to erosion from rainfall or wind.

- The closed peanut canopy also suppresses weeds to reduce weed pressure, especially when peanut is intercropped with a grain crop. In either case, reduced weed pressure reduces need for environmentally-harmful chemical weed control.

Economic growth

- Peanut provides a source of cash income for small-holder farmers, and rural and urban processors in developing countries.

- Biological N fixation by peanut reduces dependence on purchased N fertilizers.

- Often, the peanut crop is processed within the village or country that produces the crop. Thus, peanut production stimulates local food processing industries and adds value to the crop.

- Weed-controlling attributes of the peanut canopy reduce the cost needed for labor-intensive hand weeding or for chemicals to control weeds.

- Peanut is a versatile crop that provides growers with many options to spread risk. With a short growing season, peanut fits well as a cash crop within a large range of cropping systems. It may be grown as monoculture where the rainy season is short, in sequence with grain crops, or beneath a long-lived orchard crop such as banana or coconut.

Improved human health and nutrition

- Peanut has special potential as a famine prevention crop. It may be planted late in a rainy season if the previous crop has failed.

- At 25% protein and 45% oil, peanut provides an inexpensive, high-protein, high-energy food for humans and livestock. It is one of the most nutritive crops available as a complement to cereal grains.

- Peanut supplies a high-quality, healthy vegetable oil for cooking. Supply of cooking oils is often inadequate in developing countries.

CONSTRAINTS ASSOCIATED WITH PEANUT PRODUCTION, PROCESSING, AND USE

While peanut offers many benefits to growers, processors, and consumers, there are still several important constraints associated with peanut production and use. The Peanut CRSP was started in 1982 to address a set of global constraints to sustainable peanut production and use. Those constraints served as the initial basis for planning and organizing the Peanut CRSP in 1980 to 1982. Based on past Peanut CRSP accomplishments and the assessment of the External Evaluation Panel in 1989, the following constraints associated with peanut production were confirmed to be valid for the current 1990 to 1995 phase of the program.

Environmental

- Dependance on chemicals to control insect pests and diseases.

- Inadequate diversity in cultivated peanut germplasm grown in developing countries and the U.S.

Socioeconomic

- Low productivity or yield loss from disease, drought, insects, and nematodes.

- Local resource management situations may prevent efficient production and use.

- Insufficient local food supply or inadequate food technology prevents growers, processors, or consumers from exploiting the full potential of peanut because it is not considered as a primary food source.

- Information not available to potential beneficiaries of new peanut production and use technologies.

- Economic losses resulting from mycotoxin contamination.

Health and nutrition

- Health hazard from mycotoxins in contaminated peanut.

- Methods needed to incorporate peanut and peanut products into safe and nutritious processed foods.

Research capacity and tools

- Inadequate numbers of trained research and support personnel in host countries.

- Research tools using molecular biology are not readily applicable to peanut. These tools enable efficient and effective germplasm improvement with respect to resistance to pests and diseases, tolerance to abiotic stresses, seed quality and quantity, and adaptation to growing season. Thus, molecular genetic tools will enhance our capacity to overcome constraints related to environment, economic growth, and human health and nutrition.

In short, the Peanut CRSP enhances the potential of peanut as a cash crop for human food and animal feed in developing countries and the United States. The Peanut CRSP contributes to increasing rural incomes, sustained productivity of agricultural land, and improved health and nutrition of peanut consumers. Furthermore, the Peanut CRSP contributes to enhancing the research capacity of developing country institutions. Collaborative

research on peanut is producing new and improved technology that improves the well-being of people in developing countries and the United States.

THE GLOBAL PLAN

Peanut is a global crop. The crop is widely distributed and important to both developing and industrialized countries. Biological and physical constraints to peanut production are similar worldwide. Research is needed to develop technologies that relieve constraints to peanut production and use, particularly in developing countries with marginal food and protein supply.

For a global crop with global research needs, the Peanut CRSP can catalyze the collaborative efforts of researchers and administrators from diverse groups, including developing countries, U.S. institutions, international agricultural research centers, non-governmental organizations, and donor agencies. The synergism of this collaboration multiplies the impact of Peanut CRSP accomplishments.

Global thrusts

Sustainable production and delivery of food for human consumption are still primary problems in the developing countries. Based on the potential contribution of peanut to sustainable, nutritious food production, the Peanut CRSP has three global thrusts:

1. To develop sustainable agricultural production and food delivery systems that are profitable, environmentally-sound, and relieve important constraints to peanut production and use.

2. To resolve resource management situations that restrict appropriate research or diminish efficiency of systems for peanut production and use.

3. To communicate research outputs to beneficiaries in developing and industrialized countries.

Peanut CRSP benefits and beneficiaries

The benefits of Peanut CRSP collaborative research are adequate quantities of affordable, healthy, nutritious food for rural and urban consumers, increased incomes for peanut growers and processors, and environmentally-sound production and processing systems. Beyond these immediate benefits of improved peanut technologies, community and national benefits ensue from greater profitability of the agricultural sector and a healthier population.

Developing country beneficiaries

- Small-holder farmers, including rural women
- Food processors
- Rural and urban consumers

United States beneficiaries

- Peanut growers
- Processors, marketers, exporters
- Rural and urban consumers

Research strengthening

Progress toward accomplishment of our three global thrusts depends on improved capacity for collaborative research. In host country institutions, the Peanut CRSP trains research and support personnel, and provides critical research equipment. Training has emphasized degree and short-term, non-degree training at host country and U.S. institutions. In both host country and U.S. institutions, the Peanut CRSP supports research activities and facilitates communication among researchers.

Cooperation and coordination

While the Peanut CRSP is the only international program that focuses solely on peanut, there are other research organizations that conduct some peanut research. Since its inception, the Peanut CRSP has enhanced research and technology transfer through cooperative relationships with other international organizations. These relationships enhance cooperative research planning to avoid redundancy across programs, and joint conduct of workshops and symposia to communicate research results. Some organizations with which the Peanut CRSP cooperates are:

- ICRISAT the International Crops Research Institute for the Semi-Arid Tropics
- ISC the ICRISAT Sahelian Center (ISC)
- CIRAD-CA the French Center for International Agricultural Research and Development, Annual Crops Program
- IDRC the Canadian International Development and Research Centre
- ACIAR the Australian Centre for International Agricultural Research

SELECTED MAJOR ACCOMPLISHMENTS -- July 1994 through June 1995

Environmental Enhancement

- IPM researchers in the Philippines have made outstanding progress in evaluating host-plant resistance, biological control, alternative management strategies, developing economic thresholds, and pest biology. The progress seen in the host plant resistance work has developed an excellent data base evaluation under Filipino production systems. The principal investigators have taken the next important step and are testing the integration of this information into IPM programs in the various production regions. Studies investigating these IPM systems are underway in the Cagayan Valley and in Central Luzon. The data from these trials have shown that overall insect damage in IPM plots is less that in standard 'farmer managed' plots and conventional insecticide use was often avoided. Some additional studies have looked at current Seed Board recommended varieties and promising cultivars used in conjunction with classical biological control programs.
  The continuation of studies validating the use of biological control programs using Trichogramma sp. and a microbial insecticide Bacillus thuringiensis show promise for future wide-scale implementation. Biological control studies have shown practicality and profitability using an integrated program in IPM evaluation trials.

  The development of IPM control strategies continue in Thailand. This included basic insecticide evaluations to improve the timing of application to increase yield and total integrated program using host plant resistance as well as cultural and biological control methodologies. Since insecticides are relied upon quite heavily in certain areas these studies are essential. Five-years IPM work are being developed into an integrated program for Thailand and for use in Regional Training as projected in the 5-year extension of the Peanut CRSP.

- Significant variability in rosette virus reaction was found among F3 and F4 families form rosette resistant x West African adapted varieties in rosette screening trials at Niangoloka, Burkina Faso. Confirmation and repeatability of reactions are being tested in 1995. Rosette virus epidemics can cause up to 100% yield losses.

- One Philippine seedboard variety, PSB Pn 2, was released in 1994. It is locally known as UPL Pn 12 or Biyaya 12, and is a valencia peanut variety derived from a three-way cross among Philippine and North Carolina lines. PSB Pn 2 yields 13-18% more than two check varieties and has better resistance to rust and late-leafspot damage. It has high seed viability even after six months pod storage at ambient storage conditions.

- Peanut transformation technology was improved in North Carolina research, decreasing the time required to produce transgenic cell lines and improving the fertility of transgenic plants. Peanut plants transformed with the peanut stripe virus coat protein gene were tested for resistance under controlled conditions. Two lines showed reduced or delayed symptoms while others appeared to develop then lose symptoms after an initial infection period.

- Two genes for resistance to root-knot nematodes were transferred from a wild species of peanut into a cultivated type. The two genes reside near each other on the same chromosome. Located between them is a molecular marker gene that allows the presence or absence of the two resistance genes to be detected without the cumbersome test of nematode resistance.

- In North Carolina, the development of a sustainable agricultural system for peanut continues to be a major objective. Efforts to minimize preventive pesticide use and to integrate biological and cultural control practices have been expanded. The use of biological control products is showing increasing promise for insect management in peanuts These studies closely correlate to similar studies in the Philippines. The validation of forecast models for the lesser cornstalk borer and spider mites has proven to be an invaluable tool for pest management. Evaluations of pesticide interactions are increasing our understanding and provide us with an approach to reduce overall pesticide dependence.

- Thrips are the insect vector of tomato spotted wilt virus disease in peanut. In North Carolina, studies have been completed on thrips dispersal, migration, and over wintering habits which have enhanced the biological and ecological information on this pest. Newly developed procedures in 1993-94 allowed, for the first time, documentation of thrips over wintering in the soil of fields previously in peanut and second year results confirmed the first year results. These studies will dramatically improve the understanding of the vector and help to manage tomato spotted wilt virus disease. The work supports similar efforts in Thailand.

Economic Growth

- Fleur 11 seed was increased in the Senegal River Basin under irrigation and at the ISRA station near Bambey. Seed supply should be sufficient for significant grower production in the 1995 season. The variety yields 25% more than presently grown varieties.

- Breeders seed increase of the runner-type Tx896100 was initiated in Texas. The new line exceeded the common Florunner variety by over 50% at sites with severe tomato spotted wilt virus disease pressure. Agronomic traits are acceptable for commercial production. Resistance to this virus is much needed, since the virus can cause significant yield losses and is becoming more widespread.

- In Nigeria, a total of 20 tons of foundation seed of medium-duration, rosette resist and non-resistant, short-duration cultivars continued to be increased in 1994 and 1995 for distribution to farmers. These varieties will fill a much needed niche for farmers and should reduce losses to the rosette virus.

- A comprehensive package for virus control on peanut in Asia has been developed in Thailand and the publication is now in press. The basic tenants of this package incorporates plant resistance, use of virus free seed and cultural practices such as adjustment of sowing date, using high plant density, and intercropping peanut with fast growing non-host crops. In addition to use in Thailand, this package can be used in Regional training proposed for Thailand in the 5-year Peanut CRSP extension.

- In Jamaica, CARDI has been working closely with the United Farmers Multi-purpose Cooperative (UFMC). During its three-year life UFMC has used CARDI/Peanut CRSP information and assistance

in peanut production, postharvest handling, storage, and marketing. Motorized and hand threshers have been adopted, including designation of a farmer to service and maintain the threshers. The peanuts produced by the farmer's are accumulated by the co-op and marked in bulk to large and small processors. Also they solar roast peanuts and make peanut candies which are sold to schools in the area. CARDI loaned a commercial plastic bag sealer to the co-op to facilitate proper sealing and storage of processed products. UFMC is marketing its own brand of weaning food called 'Manna", which is produced by Jamaica Cereal Foods but made from peanuts and corn grown by the coop. A range of peanut flavored milk drinks are also being prepared by the co-op, which is being done in conjunction with a commercial operator that markets milk solids. UFMC is negotiating a lease on a building for office and processing space. The facility will be a major boost to the peanut producers in St. Elizabeth's Parish, which produces most of the peanuts in Jamaica.

- In Thailand, design criteria for modification of an existing sheller was developed for the Khon Kaen 60-1 variety which was developed and released with Peanut CRSP support. Improved shelling resulted from the modification, which will assist in increased farmer acceptance of the variety.

- Nutritional and microbiological quality of peanut paste from several regions of Burkina Faso was analyzed. Quality of traditionally made past was generally inferior to the product manufactured by a local industry. Results encourage the development of industrial processing plants for manufacture of peanut products. The optimum package size and material were identified by scientists at the University of Ouagadougou and information delivered to the peanut paste factory, CITEC-HUILERIE.

- Interdisciplinary and integrated efforts to transfer peanut processing technologies to rural women cooperatives in Letye, Philippines continued. One of the groups that provides assistance to the typhoon Uring victims has support of the Philippine Red Cross. Training, processing, management and marketing aspects were given increased and expanded attention. Successful production and marketing activities are in place.

- A socioeconomic study was completed in Jamaica and a written report is being developed. The survey details the peanut production, harvesting, postharvest handling, storage, processing, and marketing. Results will aid the extension service in identifying points to intervene in improving the peanut system.

- During the year several Field Days were held in Jamaica by the Ministry of Agriculture, Extension Service. In addition to information on general peanut agronomy, the Peanut CRSP developed cultivar, thresher, and sheller were featured or demonstrated.

- A survey on production and harvest technologies was conducted in Ghana. Little mechanization is used which may contribute to low production levels due to the high cost of manual labor required for production. There appears to be a need for introduction of more improved varieties, with attention given to farmers regarding their preferences for particular types. Farmers had little knowledge about the aflatoxin problem in peanuts, therefore there is an urgent need to educate and create more awareness about the problem, health effects, and ways of preventing or reducing contamination. Formation of peanut cooperatives could be an avenue for dissemination of information. Simple and appropriate technologies for peanut production need to be developed for use by Ghanian farmers or transferred from other programs and adopted in Ghana. Resolution of production problems would result in subsequent increase in production and utilization.

Improved Human Health and Nutrition

Mycotoxin reduction:

- The development of methods continued in Texas to screen for norosolenic acid, an aflatoxin precursor. Screening for norosolenic acid will allow easier evaluation of germplasm for resistance to *Aspergillus flavus* infection or aflatoxin production. Visual color differences among seed from freshly harvested peanut pods following inoculation with Aspergillus flavus mutants were strongly correlated with aflatoxin content of the seed. The norsolorinic acid mutant (red color) may give superior results that a wildtype aflatoxin producing strain of Aspergillus in analyzing aflatoxin contamination of peanuts.

- In Ghana, research has continued on evaluating promising plant extracts for control of Aspergillus infestation and/or aflatoxin production. The native plant species under study may provide an appropriate technology for control of aflatoxin production during storage. Extracts from five plant species have completely inhibited fungal growth.

- In Burkina Faso, an extract of Allium sativum was found to be an effective inhibitor to the growth of Aspergillus flavus, the fungus that produces aflatoxin. Research continues to determine the best method for application to peanut in storage. Application of this technology to peanuts in storage will have a tremendous impact on the health conditions of the local population.

- An aflatoxin survey on peanut in Ghana showed that damaged seed contained in excess of 30 ppb (WHO minimum level), while undamaged seed had generally very low levels. Hand sorting can be an effective way of reducing aflatoxin levels in seed intended for consumption.

- In Texas, the intire sterigmatocystin (ST) pathway of Aspergillus nidulans was identified. Evidence shows that the ST pathway is functionally equivalent to the aflatoxin (AF) pathway in Aspergillus flavus and Aspergillus parasiticus and that this ST/AF toxin pathway is conserved as a cluster of genes in all aflatoxin producing Aspergilli. Most importantly, the regulatory gene aflR, is found to regulate both the ST and AF pathway in all three Aspergillus Spp. Efforts will be focused on identifying ways to keep aflR turned off permanently. The study of this and other genes in the ST/AF pathway will lead to a molecular understanding of what environments, peanut metabolites, or other influences result in aflatoxin production in peanuts.

Food processing:

- In Alabama optimum conditions for the development of infant weaning foods using mixes of peanut pastes with cowpea, pearl millet, and teff flours were studied in relation to application in Burkina Faso. Good products have been make in Burkina Faso using maize, sorghum and rice, peanut flour and milk. The product has improved nutritional value (18-20% protein, 10-12% fat, 2-3% minerals) and higher energy density.

- Research is underway in Alabama to develop an improved, acceptable low-fat peanut butter. Optimum conditions (roasting, oil extraction procedures, type of fat replacers) were established for manufacture of a peanut paste with 50% less fat than commercially available products in the USA. Sensory characteristics (color, texture and taste), viscosity and storage stability (shelf life) were determined. Since flavor is associated with the oil, work is in progress to evaluate the flavor quality of the paste using Gas Chromatography techniques.

- Optimization studies for a peanut butter-honey spread were conducted at the University of Georgia. From prediction equations a formulation containing 7% dry honey and 1% salt would was acceptable in all areas except texture, which was grainy from the dry honey.

- In Georgia, a low-fat, peanut-base beverage was developed from roasted peanut. Research continued to improve the acceptability of the product. Chalky mouth feel was eliminated when partially defatted

roast peanuts are milled and wet-blended, and slurries are filtered before being pasteurized and homogenized. The product has processor interest and market potential.

- Tempeh starters for fermentation of peanut products were prepared using several substrate formulations in Thailand. Selection of starter was based on efficiency in producing good quality peanut tempeh. The starter was prepared in larger scale and tested before shipping to housewives at Kud Jub District, Udorn-thanee Province. The women are being trained to produce and market peanut products in an outreach program.

- The suitability of peanut as an extender of ground pork and beef in meatloaf was evaluated in the Philippines. Results indicate that a formula containing up to 83% defatted peanut has favorable sensory quality and equally acceptable to commercial meatloaves with similar nutritional value and physical characteristics.

- In the Philippines, supplementation and optimization of Lady Finger formulas with peanut was accomplished. Appropriate blends of peanut, wheat flour and cassava flour to give highest levels of sensory quality were defined. Packaging technologies for the baked root crop products extended with peanut were evaluated. Shelf life predictions for various conditions of packaging and storage were established. These technologies support the women's cooperative activities in processing and marketing the products.

Molecular Genetics

- In Georgia, plants are growing in the green house that have been transformed with the coat protein genes from tomato spotted wilt virus, peanut stripe virus, and peanut chlorotic streak virus. These plants either have set seed or are currently setting seed and thus are fertile All plants will be permitted to set seed and carried to the third generation where the progeny are expected to be homozygous for the transformed trait.

  Additionally, selections are being made of possible transformants that have the coat protein gene for the peanut rosette assistor virus and bud necrosis virus.

Manpower Development

- Graduate Student Training

  In U.S. Institutions

  One host country student completed a Ph.D. program with CRSP support.
  One host country student completed an M.S. program with CRSP support.

  Four U.S. student completed M.S. programs with partial CRSP support.

  One non-host country student completed a Ph.D. Program with partial CRSP support.

  Three non-host country students completed M.S. programs with partial CRSP support

In Host Country Institutions

One student completed a doctorate degree at a host country University with CRSP support.

Two students completed M.S. in host country Universities with CRSP support.

- Five U.S. collaborators traveled to host countries and provided various types of research training.
- Five Host Country collaborators have come to the U.S. for short-term training.

COMMUNICATIONS

Workshops

- In Belize, training and workshop sessions were conducted by Dr. Margaret Hinds, University of Georgia Food Technology collaborator in cooperation with Dr. Anil Sinha of CARDI. The activities included. 1) Visited three local peanut processing plants and advised them on improving the quality of their peanut products, particularly peanut butter. The current processes were analyzed and suggestions for modification developed. 2) Conducted consumer taste panels on peanut butter made in Belize and at The University from Belize grown peanuts. Results from the panel evaluations were used to further guide the processors improving peanut butter quality. 3) Held a one-day training workshop on preparation and testing of peanuts for the presence of aflatoxin. Presently only peanuts exported to the Caribbean community are analyzed for aflatoxin, which is a minor part of total production. The training increased awareness of the aflatoxin problem in locally consumed and suggested simple ways to reduce contamination.

- The Peanut CRSP sponsored a Symposium Washington, DC March 29-31, 1995 entitled "Peanut CRSP Symposium, Impacts and Scientific Advances through Collaborative Research on Peanut", The Symposium was designed to review major accomplishments of the CRSP over the life of the program with special emphasis on the period 1990-1995, and was planned in view of the CRSP closure in June 1995. All projects were represented with U.S. and Host Country principal and co-principal investigators.

- A two week training course on viruses infecting peanut and other legumes was held in Khon Kaen, Thailand during February 1995. This training was supported by the Peanut CRSP, FAO, ICRISAT and a Belgium project. It was held at Khon Kaen University with participants from 10 Southeast Asian countries. Sompone Wongkaew, Thailand and J.W. Demski, USA were the two instructors representing the Peanut CRSP.

- As part of the technology transfer activities of the Thailand Food Technology Project, Peanut CRSP researchers cooperated with home economists from the Department of Agriculture Extension, Ministry of Agriculture to transfer technology of ground roasted peanut, hot sand roasted peanut and coated peanut products to housewives at Kud-Jub District, Udorn-thanee Province. Housewives were very receptive and are producing these products for markets in their communities.

- A Workshop in Training the Trainers from 13 countries on the topic of "Quality Evaluation and Utilization of Food Legumes" was held at Kasetsart University from April 24 to May 27, 1995. The Workshop was attended by 20 participants from 13 countries: Tanzania (2), Fiji (1), Sri Lanka (1), Republic of Maklives (1), Bhutan (2), Bangladesh (2), Sierra Leone (2), Laos (2), Cambodia (1), Korea (1), Vietnam (2), Nepal (1), and Thailand (2). All participants were government officers in work is related to legume utilization. Proceedings were distributed at the Workshop. Many examples given during the training program were peanut products developed by the Peanut CRSP project in Thailand.

Cooperation with the Department of Technical and Economic Cooperation, Office of Prime Minister under the Thai-AID project facilitated the training course. This training course will be conducted annually and will result in a large impact on processing of legumes as a whole and the nutritional well-being of consumers throughout Southeast Asia as well as in other developing countries in the world in future years.

- Cooperators at the Guadalupe Women's Association and URIVAC were trained in processing, management and marketing of peanut-extended snacks by Peanut CRSP investigators at ViSCA, Letye, Philippines. Sessions were held a ViSCA as well as in rural areas in the Baybay vicinity.

- Dr. Anna Resurreccion, Peanut CRSP food science co-principal investigator, trained staff members of the Food Development Center (FDC) of the National Food Authority, Ministry of Agriculture in Taguig, Metro-Manila, Philippines on sensory evaluation methodology, specifically on descriptive analysis techniques and consumer acceptance testing procedures.

- Training courses on sensory evaluation and quality control and consumer testing of food products were conducted by Dr. Resurreccion, Peanut CRSP food science co-principal investigator, on November 28 to December 2, 1994 in the Malaysian Agricultural Research and Development Institute (MARDI) in Serdang, Selangor, Malaysia. A total of 105 (35 Philippine and 60 Malaysian) participants, mostly from the food industry and a few from government agencies and academic institutions, enrolled in the courses. The courses were sponsored by USAID (PITO Project), Technonet Asia and the FDC and MARDI in the Philippines and Malaysia, respectively.

- Dr. Larry Beuchat, University of Georgia Food Science Peanut CRSP principal Investigator participated in the 'East, Central, and Southern Africa Food Science and Technology Conference '94: Challenges for Africa Towards the Year 2000' held September 12-16, 1994 at Victoria Falls, Zimbabwe. He presented a paper entitled ' Advances in food mycology methodology'.

Special Publications and Releases (varieties, etc)

Abilay, R.M., and L.A. Lantican. 1995 PSB Pn 2 (UPL Pn 12) - A newly released valencia peanut variety at the Institute of Plant Breeding, University of the Philippines at Los Banos. Phil. J. Crop Sci. 20 (Suppl. No. 1):9.

Release of CBR-resistant NCSU breeding line N90013 E as 'NC 12C', pending approval by institutional committees and administration.

Release of UPLB breeding line IPB Pn 85 2-40 in the Philippines.

Working Together on Groundnut Virus Diseases - Transformation and Regeneration, Groundnut Viruses in Africa, and Groundnut Viruses in Asia. Summary and recommendations of a meeting of international working groups on groundnut virus diseases. 15-19 Aug 1993, Scottish Crop Research Institute, Dundee, UK. ICRISAT 1994. 81 pp. Co-sponsored by Scottish Crops Research Institute, Peanut CRSP and ICRISAT.

Groundnut Bacterial Wilt in Asia. Proceedings of the Third Working Group Meeting. 4-5 July 1994, Oil Crops Research Institute, Wuhan, China. ICRISAT, 1994, 153 pp. Publication co-sponsored by Peanut CRSP.

Mehan, V.K., C. D. Mayee, T.B. Brennenman, and D. McDonald. 1995 Stem and Pod Rots of Groundnut. Information Bulletin no.44, ICRISAT. 23 pp. Co-sponsored by Peanut CRSP and ICRISAT.

PEANUT CRSP ADMINISTRATION AND MANAGEMENT

The Peanut CRSP Management Group includes: the Management Entity at the University of Georgia with a Program Director, Assistant Program Director, Administrative Secretary, and a part-time accountant; the Board of Directors with representatives from Alabama A&M University, The University of Georgia, North Carolina State University, Texas A&M University, and ICRISAT; the Technical Committee with representatives from the above four universities and rotating representation from host countries; an External Evaluation Panel with seven members; an AID/R&D/AGR Program Manager; and a BIFADEC liaison.

Management Entity

The Management Entity is responsible to US AID and to the participating U.S. and host country institutions for fiscal and programmatic activities.

Activities:

- Provided logistical and managerial support to Investigators, including program planning, travel clearance, and equipment purchase.
- Prepared necessary reports including; Annual Report for 1994, quarterly reports, quarterly Newsletter, and Annual Work Plans and Budgets.
- Published three special articles in cooperation with ICRISAT: see special publications listed earlier.
- Arranged Board of Directors and Technical Committee Meetings.
- Arranged and participated in Annual Peanut CRSP Meeting in Tulsa, OK, July 14-15, 1994 with the American Peanut Research and Education Society Meeting July 12-15.
- Participated in CRSP Council activities to promote all CRSPs, to foster cooperation among CRSPs, and to seek inter-CRSP research opportunities.
- Coordinated CRSP contributions to the International Arachis Newsletter.
- Met with USAID Office of Agriculture and Food Security leaders, Drs. John Lewis and Harvey Hortik, in Raleigh, NC on May 9-10, 1995 to discuss plans for a Peanut CRSP five-year extension. The extension is to be guided by an Expert Panel of 8-10 people and the CRSP will be opened to any interested participant.
- Prepared plan for a one-year extension, July 1995 to June 1996.
- Prepared draft of a five-year extension proposal.

Board of Directors

The Board is the policy making group in the CRSP and is responsible for establishing the general directions for the CRSP as carried out by the Management Entity. The Board also acts on recommendations from the Technical Committee on budgets, programs, and other CRSP concerns.

Activities:

- Met in Raleigh, NC on May 9-10, 1995 to discuss a five-year extension plan with USAID officials.
- Conducted conference call meeting on June 29, 1995.

- Approved Work Plans and Budgets for 1995-96 in the June 29 Conference Call.

- Reviewed and concurred with the membership and plans for the Expert Panel that will guide the five-year extension plans (June 29 conference call).

- Dr. Onuma Okezie, Alabama A&M University is assumed the role of Chair for a two-year term. Dr. Ed Kanemasu, University of Georgia, was elected Vice-Chair.

Technical Committee

The Technical Committee is responsible for formulating program and budget plans for recommendation to the Board. They are also responsible for internal evaluations of the program and advising the Management Entity and Board of any concerns.

Activities:

- Conducted a meeting at Griffin, GA December 1 and 2, 1994.

- Communicated by Conference Call on May 17, 1995. Approved budget and plans for the one-year extension, July 1995 to June 1996, discussed plans for the five- year extension, July 1996 to June 2001, including potential members of the Expert Panel to guide the Extension Process.

- Dr. Tom Isleib, North Carolina State University is Chair and Dr. Manjeet Chinnan, The University of Georgia, is vice-Chair.

External Evaluation Panel

The External Evaluation Panel is designed to give an unbiased view of how the CRSP is progressing toward meeting its objectives. The present EEP members follow.

Dr. Bo Bengtsson, Professor in International Crop Production Science, Swedish, University of Agricultural Sciences, Uppsala, Sweden (formerly Director General of the Swedish Agency for Research Cooperation with Developing Countries and presently Chairman , CIFOR Board of Trustees), research management, crop production.

Dr. John Cherry, Director, USDA/ARS, Eastern Regional Research Center, Philadelphia, PA, food technology, research management.

Dr. Milton Coughenour, Professor of Rural Sociology, University of Kentucky, Lexington, KY, sociological change, technological change, international agricultural development.

Dr. David Hsi, Emeritus Professor of Plant Pathology, New Mexico State University, Albuquerque, NM, peanut pathology, peanut crop management systems, past-President (1982-83) American Peanut Research and Education Society.

Dr. Robert Schilling, Senior Scientist, CIRAD-CA, Montpelier, France, peanut production and use systems/West Africa, research management.

Dr. Joseph Smartt, Reader in Biology, The University of Southampton, UK, genetics of peanut, peanut production and use systems/Southern Africa.

Dr. Handy Williamson, Jr., Professor and Head of Agricultural Economics and Rural Sociology, University of Tennessee, Knoxville, TN, economic and rural development, research management, impact analysis.

- The EEP completed their intensive review in April 1995. The Report was assembled in final form in July 1995.

US AID Program Manager

The Program Manager, Dr. Dianne Janczewski, AID/R&D/AGR, interacted with the CRSP on a continuing basis, and provided the primary linkage for the CRSP with AID.

Activities:

- As an ex officio member, participated in the Board and Technical Committee Meetings.
- Provided necessary actions on travel clearances, equipment purchases, etc.
- Coordinated and promoted CRSP Council activities for the Peanut CRSP.
- Provided liaison with the CRSP for all information from AID important to the functioning of program.

BIFADEC Liaison

The liaison, Mr. William Fred Johnson, provides a link with BIFADEC for information and actions important to the Peanut CRSP.

Activities:

- Provided information regularly to the CRSP, including participating in the monthly CRSP Council Conference calls.
- Provided periodic counsel to the Management Entity related to CRSP - USAID relationships.

PEANUT CRSP IN RELATION TO EXPORT COMPETITION

Host countries of the Peanut CRSP do not compete in the edible peanut export market. Some West African countries, particularly Senegal, export peanut oil and oil meal, but the United States does not export oil or oil meal. The Senegal oil and meal export is by agreement with France and is not open to other countries on a competitive basis. The U.S. Department of Agriculture, Foreign Agricultural Service data lists minor amounts of peanut export from some host countries, for example, from Mali. This export is probably not export of edible peanut, but oil seed being shipped to a mill in another country in the Region for extraction.

PLANS FOR 1995-1996

Research and Training

Research and training in the Peanut CRSP will continue toward five major outputs: 1) reduced dependence on chemical pesticides through improved disease and pest resistant cultivars; 2) more profitable, sustainable peanut production through better adapted, higher quality, and higher yielding cultivars; 3) improved human nutrition through increased use of peanut products; 4) improved human health through prevention of exposure to mycotoxins; 5) more profitable peanut processing through improved post-harvest handling systems.

While most research in 1995-1996 will follow the plan established in 1990, there are several proposed changes to the research program.

- An Inter CRSP project to include Peanut, Soil Management, and Sorghum/Millet CRSPs will be funded in Niger by the USAID Mission to Niger.
- Research on technology transfer methods for food processing will continue in collaboration with the Visayas State College for Agriculture in Baybay, Leyte, Philippines and University of Georgia.
- The insect management project in Burkina Faso will be discontinued.

Proposal for a New Five-Year Extension

Scientists participating in the Peanut CRSP have made great progress in developing new technologies for peanut production and processing. There are, however, new and continuing problems facing peanut growers, processors, and consumers: 1) Mycotoxins are still significant health hazards; 2) There is great scope for reducing dependence on chemical pesticides; 3) Peanut products have great, untapped potential for improving human nutrition.

The planning process for the 5-year extension was put on hold in March 1994 with the announcement by USAID that the Peanut CRSP would terminate with the end of the present grant, June 30, 1995. During the course of the year this decision was reversed and the Peanut CRSP in a one-year extension from July 1995-June 1996 was charged with the development of a new five-year plan. The meeting in Raleigh, NC in May 1995 (see ME and Board activities above) launched a planning effort for a five year extension.

TX/BCP/WA
Disease Resistant Peanut Varieties forSemi-Arid Environments

Texas A&M University
Institut Senegalais de Recherches Agricoles, Senegal
University de Ouagadougou/Institut Developement Rurale, Burkina Faso
Institut Economie Rurale, Mali

O.D. Smith, Principal Investigator, TAMU; A. Ba, ISRA, Senegal; P. Sankara, UO-IDR, Burkina Faso; Kodio, O. IER, Mali; Co-Principal Investigators. N'Doye, O., ISRA; M. Sanogo, IER; C.E. Simpson, A.M. Schubert, W.J. Grichar, TAMU, Co-Investigators.

INTRODUCTION

Cultivars that will successfully and reliably produce good yields of a safe and nutritious peanut are essential to sustainable agriculture in West Africa. Irregular rainfall varied in quantity by locales and regions, soils with low moisture holding capacity, acidity and fertility, diseases, arthropods, weeds, and other factors limit production and degrade quality potential. Limited financial resources restrict grower inputs for crop production and protection. The development and utilization of improved varieties is the preferred and most effective means of improving peanut quantity and quality.

Rainfall was reasonably good in 1994 at most West African test sites both in duration and in quantity. However, yields were not as high as might have been anticipated. Early leaf spot incidence and severity was high in the more humid areas. Seedling diseases caused some loss in stand and viruses, rosette and clump, were detrimental at some sites.

In Texas, dry conditions prevailed in 1994 and dependence on irrigation was heavy. Plants in the College Station tests were distorted by pesticide contamination of the surface irrigation water source. Spotted wilt virus and sclerotinia blight pressure was heavy in areas and caused substantial economic loss but was useful for disease response evaluations. Early fall freeze and late season rains made retrieval of some germplasm difficult.

ACCOMPLISHMENTS

Breeders seed increase of runner-type Tx896100 was initiated. Single plant progenies were increased in the off-season and summer advance is in progress. Yields of Tx896100 exceeded Florunner >50% at sites with severe spotted wilt disease pressure. Shelling and quality tests indicate Tx896100 acceptable for commercial production.

Fleur 11 seed was increased in the Senegal River Basin under irrigation and at CNRA/ISRA. Seed supply should be sufficient for significant grower production.

Evaluation of both breeding procedures and progress in breeding for sclerotinia resistant runner type peanut was completed. Comparable levels of resistance was found in dense and open canopy plant forms. Greenhouse screening for sclerotinia infection was found effective in eliminating the highly susceptible plant progenies.

Significant variability in rosette virus reaction was found among F_3 and F_4 families from rosette resistant x West African adapted varieties in rosette screening trials at Niangoloka, Burkina Faso. Confirmation and repeatability of reactions are being tested in 1995.

Runner-type breeding lines from crosses of Tamspan 90 and successful runner cultivars showed good sclerotinia resistance, good plant form, good pod size and shape, and significant yield advantage over accepted commercial

varieties in yield trials with sclerotinia blight pressure. Yields and values per acre were lower, although some not significant (P=.05), in tests without disease pressure.

Spanish F_2 derived populations grown in 3 diverse Texas locations were relatively consistent one with another in oleic/linoleic composition over locations. In general, O:L ratios were lower on the Southern High Plains than in South Texas.

GOALS

Develop superior peanut lines and varieties for stable and profitable production in West Africa and in Texas.

Enhance the quality of peanut products through breeding and management.

Strengthen and increase the effectiveness of peanut improvement programs through interaction, training, encouragement, and collegial effort.

OBJECTIVES

Develop, increase, and release peanut varieties with increased stability and improved productivity in important ecological zones of West Africa and southwest U.S.A.

Incorporate quality oil traits into improved lines with resistance to sclerotinia blight, TSWV, and leaf spot.

Assess the potential usefulness of various candivars and cultivars in Sahelian West Africa and identify production constraints which must be addressed in the development of successful varieties for the region.

Enlarge and enhance the germplasm resources of the collaborative peanut improvement programs.

Develop improved methods of evaluating disease resistance.

Assess chemical quality, aflatoxin suppression, and arthropod response characteristics in the evaluation of candivars.

Encourage participants and increase the expertise of host country scientists in the development and culture of improved peanut varieties.

APPROACH

Field screen and select among populations and breeding lines derived from short-growth duration and adapted parentage for identification and verification of useful, short growth duration, agronomically desirable genotypes.

Develop by hybridization and pre-select in Texas within and among populations with early maturity, disease resistance (leaf spot, spotted wilt virus, rosette virus, rust, soil borne diseases), high quality, and productive parentage for adaptation and utility in collaborating country programs.

Interact and collaborate with other CRSP projects, especially those active in West Africa, to increase research effectiveness and gain needed information on quality factors, including aflatoxin production, of elite breeding lines and germplasm, soil characteristics in relation to differential variety performance and/or cultural management, and disease and arthropod resistance.

Cooperate with ICRISAT, CIRAD-CA, and other regional programs in addressing the important constraints to peanut production in West Africa.

Evaluate the disease reactions of select germplasm lines and checks in Burkina Faso with relatively high rainfall where disease pressure is conducive to effective evaluation and screening.

Assist and encourage host country scientists in achieving project goals and developing effective peanut research programs.

ORGANIZATION AND PERSONNEL

U.S. Lead Institution	Texas A&M University (TAMU)
Principal Investigator	Dr. Olin D. Smith, Department of Soil and Crop Sciences (S & CS), TAMU, College Station
Co-Investigators	Dr Charles E. Simpson, TAMU Research and Extension Center, Stephenville
	Dr. A. M. Schubert, TAMU Research and Extension Center, Lubbock
	Mr. W. J. Grichar, TAMU Research & Extension Center, Yoakum
Cooperators	Dr. Nancy Keller, Department of Plant Pathology & Microbiology. TAMU, CS
Research Associate	Mr. Brent Bessler, TAMU Agri. Research Station, Yoakum
Research Assistants	Mr.Mahama Ouedraogo, Dept. of S & CS, TAMU, CS
	Mr. Michael Baring, Dept. of S & CS, TAMU, CS
	Mr. Gary Thomas, TAMU Res. and Ext. Ctr., Stephenville
	Ms. Yolanda López, Dept. of S & CS, TAMU, CS
	Mr. Jason Goldman, Dept. of S & CS, TAMU, CS
	Mr. Patrick Pace. Dept. of S & CS, TAMU, CS
Budget Analyst	Ms. Judy Owens, TAMU Research Foundation, CS
Secretary	Ms. Anja Boykin, Dept. of S & CS, TAMU, SC
Institutional	Dr. Dudley T. Smith, Associate Director, Representatives Texas Agriculture Experiment Station, TAMU, CS
	Dr. E.C.A. Runge, Head, Dept. of S & CS, TAMU, CS

RESEARCH ACCOMPLISHMENT

Yield Trials

Tests were conducted in the country of each participant but test entries varied among and within countries. Cooperators grow new accessions two or more years at their primary locations and select entries for the more advanced and multiple location trials.

Burkina Faso. Thirty-two entries including 3 check cultivars were compared at three locations: Tenkodogo, Gampela, Bobo Dioulasso, and Niangoloka. Plant stands at harvest were relatively low and this was reflected in yields. Particularly poor were the harvested stands at Niangoloka and those at Bobo Dioulasso were only marginally better. As a result, yield differences among entries were not significant. Seven of the entries, including three checks, had been evaluated for three years and 16 for two years. Yields fluctuated markedly among years and both average pod yield and stability of yield are of concern. Yields of the checks were consistently among the better entries in average yield each year. At Tenkodogo, two of the lines averaged higher in yield for years 1992-1994 than the mean of the three checks but none were superior to the best check. Two entries evaluated for only two years exceeded the best check by 10%. However, the yield advantage was not

constant over the two years. Three breeding lines yielded well compared to the majority of the entries for the multiple year periods and should be tested further for examination of stability of production over years. Plant stands at harvest, particularly at the higher rainfall sites, are a concern. Average stands at each of the four locations was less than 50% in two of the last four years of testing.

Senegal. A total of 32 and 45 breeding lines plus checks were evaluated for yield and other traits at Bambey and Nioro, respectively. The entries were divided into 3 balanced lattice tests at Bambey and five at Nioro. Entries differed between locations because of length of growing seasons. At Bambey, clump virus was apparent and ranged in incidence from 1 to 16% at harvest. Plant stands were generally good although charcoal rot caused some plant death. Fleur 11, used as a check, continued to yield exceptionally in all three Bambey tests, and was not equaled by any of the other entries for pod yield.

At Nioro, plant stands varied among entries and ranged from 50 to 88% at 40 days after planting (DAP). The poorest stands were in the large-seed confectionery test where crown rot caused an average 14% reduction in plant stand (from 74 to 60%) based on counts made 20 and 40 DAP. Early leaf spot was prevalent with severities 105 DAP ranging among entries from 5.7 to 8.7 as rated by the ICRISAT scale. Pod yields for none of the lines in the oil test were superior to the check. Two of the confectionery lines showed some promise in yield but the %SMK of all entries was very low with an average of 35%.

Mali. Thirty lines from Texas from crosses of 55-437 and Chico were compared in a randomized complete block design with three replications at Samé. The objective was to identify one or more short season lines with drought tolerance. The test was planted July 24 and harvested October 22 (90 days). Data collected included numbers of plants emerged and survived until harvest, days to flowering, weight of 100 pods and 100 seeds, haulm and pod yield, and notations on leaf spot if important. Pod yields ranged among entries from 700 to 1715 kg/ha. The pod yield of thirteen lines was equal to or better than the 47-10 check. Two lines yielded haulms of 400 kg/ha or more. Selections with both high pod and haulm were retained for further testing. The shelling percentages of almost one-third of the entries was 75% or greater at the 90 day harvest which is indicative of early maturation. Leaf spot incidence and severity was moderate and 59% scored 3 or less by the ICRISAT scale.

Disease Resistance

Leaf Spot and Rust.

Burkina Faso. Leafspot was heavy on tests at Niangoloka and Bobo Dioulasso. Late season disease ratings, ICRISAT scale, for 30 lines plus three checks at Bobo ranged from 4 to 9 with RMP-12 receiving a score of 5 and TS 32-1 an 8. Leafspot scores at Niangoloka were similar with range of 3 to 9 and 3 and 8 for the two checks.

Rust development was also evident in the southwestern part of the country. Rust scores for the 30 lines ranged from 1-3 at Bobo and 2-5
Niangoloka. RMP-12 was scored a 5 at Niangoloka and TS 32-1 a 3.

Mali. Twenty-four ICRISAT lines and 28-206 were compared at Kita for yield grade, fruit size and leaf spot reaction. Disease severity was not strong as ratings by the ICRISAT scale ranged from 2 to 4. None of the lines were superior in disease reaction to the check. Up to a 20% yield advantage was recorded for some lines.

Texas. Twenty-six selected $F_{2:4}$ progenies from intercrosses of selected interspecific germplasms were compared for yield, grade and disease reactions at Yoakum with and without fungicide protection for agronomy and leaf spot evaluation. Yields of the best selections approached that of checks Florunner and Southern Runner and leafspot reactions of some entries were equal or better than Southern Runner. Eight populations were chosen for re-selection.

Seven hundred four BC_2F_4 and 318 BC_2F_3 interspecific derived populations were planted at Yoakum for natural early leaf spot infestation. Disease pressure was moderate to weak. Seventy populations were selected for continuation based on disease reaction and agronomic traits.

RAPD analyses on leaf spot resistant parental and backcrosses interspecific progenies revealed a higher incidence of polymorphism between *A. hypogaea* and *A. cardenasii* Kaprov and W.C. Gregory and *A. diogoi* Hoehne respectively, than between *A. hypogaea* and the advanced interspecific lines. The results indicated that DNA segments were successfully transferred from the wild species to the cultivated *A. hypogaea* and that genetic diversity in peanut can be assessed using RAPD markers. Evidence of association between any of these RAPD markers to early leafspot resistance or any trait of interest was not established.

Eight F_2 populations from crosses of recently collected leaf spot resistant hypogaea x adapted peanut populations were planted at Yoakum for yield, grade, and leaf spot evaluation. Six populations were selected having small to good runner pods size and good leaf spot reaction from which to make reselections.

Ninety seven second-cycle intercross interspecific F_2 runner-type progenies were planted at Yoakum for selection and initiation of crosses for third cycle .

Sclerotinia blight

Favorable reports on partially resistant, spanish variety Tamspan 90 were frequent in North Texas and Oklahoma, but the need is strong for a good, industry acceptable runner variety with sclerotinia resistance. Disease incidence was high in 1994 as conditions favored disease development. Forty-eight lines selected on the basis of disease reaction, plant form, and pod and seed characters were evaluated for yield, grade, and other factors at sites with varied sclerotinia pressure in 1994. Entries were selected based on disease reaction, vine form, and pod traits in earlier tests. Entry numbers were varied among sites because of seed availability. Seed only from sclerotinia free fields were used for planting tests on non-infested land to avoid risk of spread of the disease.

The greatest amount of testing was in Central Texas but several were lines were compared in Caddo County, Oklahoma in cooperation with Dr. Hassan Melouk, USDA-ARS, Stillwater, and in South Texas for evaluations on adaptation and reaction to spotted wilt disease. Most tests were comprised of four replications and all entries within a test were harvested simultaneously. Yields of breeding lines Tx901417 and Tx901269 were much higher than Florunner in Central Texas and Ft. Cobb, Oklahoma where sclerotinia blight was severe. In South Texas these lines yielded less than Florunner in the presence of spotted wilt virus. Tx901305 showed some promise at Ft. Cobb and in South Texas. Florunner was superior in Central Texas in the absence of both sclerotinia blight and spotted wilt virus.

Twenty-eight of the entries were compared with checks in two Central Texas tests without disease pressure and in one test with disease. However, rain prolonged digging of the test with disease and caused some confounding of data. Yields of some entries were statistically equal to the three commercial checks but most were lower. Grades and seed size of most entries were not different from Florunner.

Another 20 entries were compared at Stephenville in two tests; one test (2A) with moderate and the other with heavy (test 2B) disease pressure. Yields, grades, seed size, and disease scores were very favorable for some entries, e.g. Tx901338-2 yielded 5244 lbs. of pods/ac compared to Florunner with 2718 pods/ac in test 2A and 4250 and 1474 pods/ac, respectively, in test 2B. The same line also was rated 1.0, in scale 1.0 = minimal to 10 = destruction by disease, whereas Florunner was 5.7 in test 2B. Four entries from this group were selected for evaluations in 1995 South, Central, and Northwest Texas and at three sites in Oklahoma.

Several runner-type breeding lines have now been selected that have yielded markedly superior to current cultivars with heavy sclerotinia disease, and equal to the cultivars without disease. Expanded testing in both Texas and Oklahoma is in progress.

Southern Blight

Twenty-four lines and checks were for a third year compared for yield, grade and other traits including southern blight reaction at Yoakum in a field with natural and enhanced inoculum. Tamspan 90 had the least southern blight "hits" among the spanish. Tx896100 was the best among the runner. A similar study of 12 entries comprised of relatively new runner-type varieties, checks, and two breeding lines, were compared at three fungicide levels was initiated in 1995.

Nematodes

Seed from the 5th backcross of nematode resistant amphiploid and U.S. adapted spanish, runner, and virginia-type cultivars and advanced breeding lines were screened for rootknot nematode reaction and seed increase was effected. Preparations were made for accomplishing the 6th backcross this fall.

Rosette Virus

Virus reaction tests was conducted on 120 $F_{2:4}$ progenies of resistant and susceptible $_{2:3}F$ families for confirmation of results and selection of superior progenies. Differences were found and data analyses and interpretation are pending.

Spotted Wilt Virus

Virus incidence, yield, grade, and other data were collected on 74 breeding lines, plus 14 checks and new varieties, in 4-replicate tests under natural in Frio County, TX. Virus readings were taken on four dates during the season by Dr. Mark Black, cooperating Extension Plant Pathologist, at Uvalde, TX. End of season disease incidence ranged from 12 to 83%. Yields of susceptible entries and checks were suppressed severely. Five lines with unit acre returns more than 30% over checks were selected for multi-location trials. Grade, growth habit, preliminary shelling, and other data were collected.

Peanut Quality

Eighteen $F_{2:4}$ spanish families with varied (1.1 to 20:1) F_3 O:L ratios were compared in South, Southeast and Northwest Texas. The major purpose was to ascertain the stability of the O:L ratios in the varied climates. The populations derived from crosses of spanish varieties x unadapted, unusually high oleic acid, spanish-type parents. The results indicated marked improvement at all locations in the oil composition as a result of the high oleic character. O:L ratios for the selections, averaged over the three tests, ranged from 1.1:1 to 19.4:1. Ratios were higher in South than in Northwest Texas. Yields and values per acre of the selections were, in general, lower than for the check varieties, Tamspan 90 and Spanco. With few exceptions, families with high O:L ratios tended to yield lower than those with lower ratios. The test, with minor additions, was planted again in 1995. Additionally, individual plant harvest was made of progenies of families with high and intermediate O/L ratios. Seed from most plants were both analyzed for O/L ratios and planted in the field. Oil composition and agronomic data are being collected.

Other studies

Germplasm Characterization

Characterization of the Malian peanut collection was continued at the Kita Station with comparative emphasis on date of flowering, growth duration, number of mature pods at maturity, leafspot reaction and plot yield. The tests consisted of 85 long (120 day), 25 intermediate (105 day), and 16 short (90 day) duration ecotypes.

Soil Amendment Study

A fourth and final year of the soil amendment study was conducted at the Farakoba Station near Bobo Dioulasso. This study, collaborative with Soils Management CRSP scientists, evaluated the use of four soil amendments on peanut production. Each year the test was conducted on the same field site using the same plot layout. Plant stands again were less than desired but better than in 1993. Again, the application of ash (2 T/ha) resulted in significant (P=.05) increase in pod yield. No benefit was realized from the other treatments which consisted of $CaSO_4$ (600 kg/ha), P_2O_5 (50 kg/ha), and $CaSO_4$ and P_2O_5 combined. The stand of RMP 12 was poorer, as in previous years, than the other cultivars (QH243C and TS32-1); however, the yields of the three varieties were not different (P=.05).

TRAINING

Name	Sex	Univ.	Degree	Date	Support
Ouedraogo, Mahama	M	TAMU	PhD	May 1995	Total
Lopéz, Yolanda	F	TAMU	PhD	Aug 1997	Partial
Pace, Patrick	M	TAMU	PhD	May 1997	Partial
Tuggle, Justin	M	TAMU	PhD	May 1997	Partial

PRESENTATIONS AND CONFERENCE PARTICIPATION

Besler, B.A., W.J. Grichar, and O.D. Smith. 1995 Germination of selected peanut varieties in small grain residue extracts. Twenty-seventh Annual Meeting of APRES. Charlotte, NC.
Simpson, C.E., D.L. Higgins, and W.H. Higgins. 1995. Long term storage of Arachis seed. Twenty-seventh Annual Meeting of APRES. Charlotte, NC

N'Doye, O. 1995. Peanut CRSP Workshop. Washington D.C. Mar 1995
Sankara, P. 1995. Peanut CRSP Workshop. Washington D.C. Mar 1995
Smith, O.D. 1995. Peanut CRSP Workshop. Washington D.C. Mar 1995
Simpson, C.E., 1995. Peanut CRSP Workshop. Washington D.C. Mar 1995
Sanogo, M., 1995. Peanut CRSP Workshop. Washington D.C., Mar 1995
Ouedraogo, M., 1995. Peanut CRSP Workshop. Washington D.C., Mar 1995
N'Doye,O. 1995. InterCrsp Workshop on National Resource Management. Niamey, Niger, Sep 1995
Sankara,P. 1995.InterCrsp Workshop on National Resource Management. Niamey, Niger, Sep 1995
Sanogo, M., 1995. InterCrsp Workshop on National Resource Management. Niamey, Niger, Sep 1995
Smith, O.D. 1995. Twenty-seventh Annual Meeting of APRES, Charlotte, NC
Lopez, Y. 1995 Twenty-seventh Annual Meeting of APRES, Charlotte, N.C.

PUBLICATIONS

Lopéz, Y. 1994. A comparison of Tamspan 90 peanut component lines for aflatoxin production. M.S. Thesis. Texas A&M University. p. 72

Ouedraogo, M. 1995. Yield and leafspot resistance of interspecific peanut crosses in early generation tests. Ph.D. Dissert. Texas A&M University. p 161

Goldman, J.J. 1995. An analysis of progress in breeding sclerotinia blight resistant runner-type peanut. M.S. Thesis. Texas A&M University. p. 39

Ouedraogo, M., O.D. Smith. C.E. Simpson, and B.A. Besler. 1995.
Combining ability for components of resistance to early leaf spot and yield of inter- and intraspecific peanut lines. OCL 2:149-152.

Taber, R.A., O.D. Smith, D.H. Smith, R.E. Pettit, J.S. Neck, S. Rajapakse, and W.F. Harman. 1994. Growth response of peanut to field inoculation with endomycorrhizal fungi, *Bradyrhizobium*, and supplemental phosphorus in Texas. Peanut Sci. 21:104-112.

Goldman, J.J., O.D. Smith, C.E. Simpson, and H.A. Melouk. 1995. Progress in breeding sclerotinia blight resistant runner-type peanut. Peanut Sci. 22: (accepted)

Smith, O.D. and C.E. Simpson. 1995. Selection of Peanut Varieties. American Phytopathological Society.

PLANS FOR 1995-1996

Senegal

a. Cooperatively evaluate lines selected as elite by project co-investigators in Burkina Faso, Mali, and Senegal.

b. Continue selection and evaluation among lines selected for dormancy among progenies of crosses with 73-30 and widely grown,non-dormant Senegalese varieties.
c. Conduct on-farm trials of PC9-79 and H75-0 to ascertain merit or release as varieties.
d. Continue evaluation of germplasm and segregating populations for leafspot resistance at Nioro.
e. Increase seed and evaluate leaf spot reaction and agronomic adaptation of interspecific germplasm lines developed and selected for early leaf spot resistance and acceptable runner traits in Texas.

Mali

a. Cooperatively evaluate lines selected as elite by project co-investigators in Burkina Faso, Mali, and Senegal.

b. Further evaluate the potential of the 13 55-437 x Chico selections from Texas for productivity and stability under drought stress conditions.
c. Further evaluate utility of Malian land varieties in varied ecological regions.

Burkina Faso

a. Cooperatively evaluate lines selected as elite by project co-investigators in Burkina Faso, Mali, and Senegal.

b. Continue evaluation of selected breeding lines at Tenkodogo, Gampela, Bobo Dioulasso, and Niangoloka for adaptation, productivity, and reaction to biological constraints.
c. Evaluate selected interspecific breeding lines selected for productivity and early leaf spot reaction in Texas for disease reaction and adaptability to Southwest Burkina Faso.
d. Further evaluate selected breeding lines for rosette reaction and adaptation.

U.S.

a. Prepare release proposal and continue seed increase of Tx896100.
b. Complete necessary evaluations of Tx896100 at multiple locations in Texas.
c. Evaluate performance of elite spotted wilt resistant and sclerotinia resistant runner type lines at multiple locations in Texas with and without disease pressure.
d. Continue evaluation and selection for improved spotted wilt disease resistance among hybrid derived families and populations in South Texas.
e. Further evaluate reaction to sclerotinia blight of approximately 400 families and lines of vaied generations for disease resistance and agronomic acceptability.
f. Select and begin evaluation of early leaf spot reaction,agronomic performance, and shelling characteristics of numerous plant selections from first cycle intercrosses of select F_2 derived populations from interspecific hybrids.
g. Select among 18 very early spanish x adapted West African and U.S. variety populations for early maturing plants for plant row evaluations in Texas and West AFrica.
h. Select plants from interspecific crosses with desirable plant form and leaf spot and spotted wilt virus reaction for plant row evaluations in Texas and West Africa with bunch and spreading plant forms from interspecific crosses
i. Select among 70 $BC_2F_{2:4\&5}$ runner interspecific runner-type progenies for agronomic acceptability and early leaf spot reaction.

j. Continue evaluation of the 12 runner-type lines and varieties for southern blight reaction with and without Tilt[R] fungicide.
k. Continue evaluation of selected spanish lines with varied O/L ratios for adaptability and stability of O/L ratio in diverse Texas environments.

Annual Report, FY1994-95

NCS/BCP/TP
Peanut Varietal Improvement for Thailand and the Philippines

North Carolina State University
University of the Philippines at Los Baños
Thailand Department of Agriculture
Khon Kaen University
Kasetsart University

Thomas G. Isleib, Principal Investigator, NCSU
Aran Patanothai, Principal Investigator, KKU, Thailand
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INTRODUCTION

Improved peanut cultivars are essential in an integrated plan to augment yields and profitability of peanut for farmers in Thailand, the Philippines and North Carolina. Crop environments and cultural practices used in peanut production vary greatly among and within peanut-producing areas. In Thailand, peanuts can be produced during a brief period of the rainy season before rice planting, after rice harvest using residual soil moisture, or in upland field conditions during the rice-producing rainy season. Irrigation may also allow some farmers to produce peanuts during the dry season. Similar conditions are present in the Philippines where the duration of the rainy season can vary between three and six months over areas where peanuts are grown. The most productive growing season in the Philippines continues to be after rice, but breeding efforts are needed for peanut to fit in other cropping systems including the shaded floors of coconut groves and upland areas with acid soils.

Constraints to production vary from place to place and system to system. In almost any situation, foliar fungal pathogens, soil-borne pathogens, and arthropod pests would destroy the peanut crop if uncontrolled. The highly capital-, chemical-, and energy-intensive production system used to achieve control in the United States is not practicable nor desirable for growers in Thailand or the Philippines where integrated programs of disease and pest management must be used. The prevailing regulatory philosophy in the United States emphasizes conservation, food safety, and environmental protection, leading to reduced use of pesticides, leading to more emphasis on the integrated approach. Host plant resistance to economically important pests must be a salient feature of this approach.

ACCOMPLISHMENTS

Philippines

One Philippine seedboard variety, PSB Pn 2, was released in 1994. PSB Pn 2 is locally known as UPL Pn 12 or Biyaya 12. It is a valencia peanut variety derived from the three-way cross between (UPL Pn 2 x FESR 1) x NC 6. It exceeded the seed yield of BPI Pn 2 by 18% and PSB Pn 1 (released in 1992) by 13% in 12 tests in the national cooperative yield trials during two dry season croppings across seven locations. It has better resistance to peanut rust and late leafspot diseases than both check varieties. Similar to PSB Pn 1, it has resistance to leafhopper damage. It has moderate resistance to *Aspergillus flavus* invasion using dry seed resistance test, and it has high seed viability even after 6 months pod storage at ambient storage condition.

Lines with high seed viability were identified 8 months after storage as seeds under conditions where no $CaCl_2$ was used.

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A total of five new entries were included in the national cooperative tests across nine locations in the Philippines in 1995. These new entries included IPB Pn 88 19-6, IPB Pn 87 24-28, IPB Pn 88 19-45, IPB Pn87-65BC11, and IPB Pn87-66BC13.

Thailand

Activities in 1994 were concentrated on yield testing of various groups of materials at different evaluation stages. Breeding lines selected for the various breeding objectives were initially tested in the preliminary yield trials. At the preliminary yield trial stage, the lines were grouped according to the individual breeding objectives and tested separately. At the standard, regional, and farm trial stages, the lines were merged into three series of trials: a medium-seeded type, large-seeded type, and early maturity lines. Promising lines from each trial were selected for entry into the next stage of testing in order to evaluate their adaptation and stability in diverse environments. All trials except the farm trials were conducted at the research centers and experiment stations of the Field Crops research Institute. Tests were conducted under rainfed conditions in the rainy season and under irrigation in the dry season.

United States

Additional data were obtained to support the proposed release of Cylindrocladium black rot-resistant breeding line N90013E.

Several genotypes were identified with resistance to early leafspot (*Cercospora arachidicola*) that was stable across day/night temperature regimes, making them useful for tropical, subtropical, and warm temperate growing conditions. Soil amendment with wheat straw was found to increase incidence and severity of Cylindrocladium black rot and southern stem rot (*Sclerotium rolfsii*) while suppressing Sclerotinia blight.

Peanut transformation technology was improved, decreasing the time required to produce transgenic cell lines and improving the fertility of transgenic plants. Peanut plants transformed with the peanut stripe virus (PStV) coat protein gene were tested for resistance under controlled conditions. Two lines showed reduced or delayed symptoms while others appeared to develop then lose symptoms after an initial infection period.

Two genes for resistance to root-knot nematodes (*Meloidogyne arenaria*) were transferred from the wild species *Arachis cardenasii* into tetraploid peanut breeding populations. The two genes reside near each other on the same chromosome. Located between them is a molecular marker gene that allows the presence or absence of the two resistance genes to be detected without the cumbersome test of nematode resistance.

GOALS

To increase yields and quality of peanut in the U.S. and cooperating Asian countries while decreasing the dependency on chemical inputs. This should improve not only the economic situation for farmers but also have a positive impact on the ecology and sustainability of agriculture. Breeding efforts need to be directed toward development of cultivars that fit current and future cropping systems and marketing potential.

OBJECTIVES

North Carolina

1. Aid in the identification of useful genotypes and implementation of material in crossing programs for use in the Philippines and Thailand.

2. Obtain greater understanding of the interrelationships between peanut host and pathogen or environmental stress that occurs in peanut-growing areas of the Philippines, Thailand, and the United States.

3. Improve breeding and selection techniques. Break barriers prohibiting incorporation of desired traits into adapted material via interspecific crossing and molecular engineering techniques. Improve screening techniques for resistance to pests and tolerance to stress.

Thailand

1. To develop peanut cultivars that can be established in specific cropping systems and seasons. Cultivars with high yields, early maturity, large seeds, resistance to rust, leafspots, *Aspergillus flavus*, *Aspergillus* crown rot, *Sclerotinia* stem rot, TSWV, stripe virus, and insects and drought tolerance would be incorporated for specific growing seasons and cropping environments.

2. Provide information on plant pathology required for development and utilization of disease-resistant cultivars.

3. Develop cultivars for the boiling-type market.

Philippines

1. To develop stable peanut cultivars that are high yielding, resistant to major foliar diseases (rust and late leafspot [*Cercospora personatum*]), leafhopper damage, longer seed viability for specific growing environments - upland optimum growing conditions, partial shaded and highly acidic soils.

2. To identify peanut genotypes tolerant to intercropping under coconut and on highly acidic soils.

ORGANIZATION AND PERSONNEL

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Mr. Donald Parry, Dept. of Crop Science
Ms. Gerry Phillips, Dept. of Crop Science
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	Quezon Agricultural Experiment Station (QAES)
	Philippine Coconut Authority (PCA) at Bago Oshiro, Davao City
	Mayors Office and Department of Agriculture at Guagua, Pampanga
	Aurora Integrated Area Development Project (AIADP)
C. USAID Project Officers:	Mr. Robert Resseguie,USAID, Manila
	Mr. Douglas Clark, USAID, Bangkok

APPROACH

Peanut germplasm is being introduced to Thailand and the Philippines. Observations on agronomic potential, disease and insect resistance, maturity, drought tolerance and other agronomic traits of the introduced germplasm are made in unreplicated nurseries. Selected lines are grown in preliminary replicated tests to identify lines for further testing at multiple locations within each country and then to on-farm trials. In addition to identifying lines for potential release as new cultivars, the tests identify parents for hybridization programs.

Crosses between germplasm with desired traits and locally adapted cultivars are made to transfer desirable traits to adapted germplasm. Pedigree, backcross and bulk breeding procedures are being used to develop improved cultivars. Hybrid populations appropriate to the environments of Thailand and the Philippines are being developed at NCSU and in both countries. Late generation material is evaluated in both countries for potential use in cropping systems peculiar to Southeast Asia such as acid upland soils, shaded floors of tree-crop plantations, or growth in paddies on residual moisture following harvest of a preceding rice crop. Promising breeding lines are tested at multiple locations in coordinated yield trials by the DOA in Thailand and by institutions cooperating with IPB in the Philippines.

In addition to exploiting the natural genetic variation within the cultivated species in the US and the Host Countries, novel genetic information is brought into *A. hypogaea* in two ways: interspecific hybridization to introgress desirable characters from the wild species, and genetic transformation using microprojectile bombardment or *Agrobacterium tumefaciens*. In utilizing the wild diploid species, barriers of ploidy are broken by chromosome doubling either before or after hybridization, then hybrids are backcrossed to cultivated parents. Transformation is being used to achieve resistance to peanut stripe virus by incorporating genes encoding viral capsid protein genes.

Short visits to both Thailand and the Philippines have been made as needed by the principal investigators to review progress, redefine objectives, plan for the next year and provide technical assistance. Short-term visits of Thai and Filipino collaborators to NCSU, ICRISAT or collaborating countries will be made as needed. Both degree and short-term training will be provided based on need and available funding.

RESEARCH ACCOMPLISHMENTS IN DETAIL

I. U.S.

A. Breeding

1. Select intensively for large-seeded adapted lines with resistance to Cylindrocladium black rot (CBR) caused by *Cylindrocladium parasiticum*.

a. In 1994, three lines with superior resistance to CBR were crossed in a factorial mating with four agronomically superior lines exhibiting intermediate resistance. F_1 seeds were sent to the winter nursery at the USDA Tropical Agriculture Research Station at Mayaguez, Puerto Rico. F_2 populations were planted in a naturally infested field in Bertie County, NC, in 1995.

b. F_2 populations from a partial factorial mating of nine sources of resistance to CBR with three breeding lines possessing superior seed attributes and high yield were screened for resistance in the infested area. Sixty-eight selected $F_{2:3}$ families were sent for increase to the winter nursery. $F_{2:4}$ families were planted in the infested area in 1995.

c. Selection for resistance and pod attributes was practiced on infested soil among and within $F_{2:4}$ families derived from 124 F_2 plants selected during the 1993 growing season. $_{4:5}F$ progenies were planted in a replicated test on infested soil in 1995.

d. 62 $F_{4:5}$ families and checks were evaluated on infested soil for CBR incidence in a completely random design with unequal replication to allow for variable seed number. These families were also increased on uninfested soil at the NCDA Peanut Belt Research Station at Lewiston, NC. Resistant families were screened for pod and seed characters, leaving only 37 which were planted in two tests in 1995: one on infested soil to re-evaluate resistance in the CBR Advanced Test and another on uninfested soil at Lewiston.

e. 34 F_4- and F_6-derived families selected were compared on infested soil with checks for CBR resistance in a replicated test and for yield and market grade at Lewiston in 1994. Nine of the lines were previously evaluated in the CBR Advanced Test in 1993; six were included in the Advanced Yield Test leading to cultivar release. Also included in the test were ten lines derived from a cross between CBR-susceptible cultivars NC 7 and NC 9. These ten had been found to exhibit partial resistance to CBR. Two lines, N93107C and N93112C were found to combine CBR resistance with superior yield, pod and seed characteristics.

f. Five CBR-resistant lines selected from the 1993-94 CBR Advanced Test were mated in diallel fashion along with six other lines selected on the basis of leafspot resistance or agrononic performance F_1 seeds will be sent to the 1995-96 Puerto Rico Winter Nursery for seed increase.

2. Selection for resistance to early leafspot (*Cercospora arachidicola*).

a. 31 F_4- and F_5-derived families from a program of selection for resistance to early leafspot were evaluated for defoliation, yield, and market grades in replicated tests at Lewiston in the 1994 growing season. Two reps were sprayed with fungicides to control leafspot and two were left unsprayed to provide an estimate of yield reduction due to disease. Several lines derived from the crosses GP-NC 343 / NC 6 and GP-NC 343 / NC 9 combined intermediate levels of leafspot resistance with superior yield, pod and seed characteristics.

b. $F_{2:3}$ families derived from 69 leafspot-resistant F_2 plants (selected on the basis of defoliation) were grown without chemical control of leafspot to permit selection for resistance among and within families. 85 F_3 plants were selected and their $F_{3:4}$ progenies planted for further selection in the absence of chemical control of leafspot.

c. F_1 hybrids derived from intermating nine early leafspot-resistant selections from NCSU with five University of Florida selections resistant to late leafspot (*Phaeoisariopsis personata*) were grown for seed increase in the field at Lewiston. Chemical control of leafspot was used to maximize seed production. F_2 populations were planted in the field in 1995.

d. In 1994, three lines selected for resistance to early leafspot and two susceptible lines with excellent yield and grade characteristics were mated as females with two highly resistant tetraploid lines developed by H.T. Stalker through introgression of germplasm from *A.*

cardenasii into *A. hypogaea*. F_1 plants were used as males in crossing back to the less-resistant parent in 1995.

3. Selection of early-maturing large-seeded virginia lines with good storability.
   a. Populations have been developed from crosses of virginia cultivars with F435, a spanish line with an extremely high ratio of oleic to linoleic acid in the fatty acid profile of the oil. F_4-derived lines homozygous for the two recessive genes conferring the trait were backcrossed to their respective adapted parents. F_1 hybrids were grown in the greenhouse during the winter of 1994-95. Progeny were evaluated for fatty acid composition as individual seeds. Seeds with high oleate content were grown in the greenhouse to be crossed back to their respective ancestral virginia cultivars.
   b. 27 advanced F_5- and F_6-derived selections from crosses of NC 7 with NC 9 and Florigiant were evaluated for performance at early (September 15) and conventional (October 1-7) digging dates. Eighteen were retained for further testing at NCSU and in the VPI/NCSU cooperative Early Maturity test conducted by R.W. Mozingo in Dinwiddie County, Virginia.

4. Incorporation of arthropod pest resistance into *A. hypogaea*. F_4 progenies were grown of crosses between large-seeded lines and a set of arthropod-resistant selections including tetraploid lines derived from an interspecific cross of *A. hypogaea* PI 261942-3 with *A. cardenasii* 10017. Single-plant selection was performed and 33 $F_{4:5}$ populations were planted in 1995 as part of a program of inbred line selection.

   The insect resistance detected to date in the NCSU peanut breeding program has derived in large part from PI 121067 through its descendant, GP-NC 343. GP-NC 343 is the source of the partial insect and leafspot resistance in NC 6, the most resistant cultivar ever to have been released to growers. Thirty advanced leafspot lines and the 35 $F_{4:5}$ selections descended from GP-NC 343 were planted in replicated tests without insecticide treatments in 1995 to evaluate their resistance to the insect complex of North Carolina. Five of the advanced lines with the largest seed supply were also sent to D.A. Herbert at VPI's Tidewater research and Extension Center for *in vitro* evaluation of resistance to Southern corn rootworm (*Diabrotica undecimpunctata*), the most damaging pod-feeding insect in the Virginia-Carolina peanut production area.

5. Recurrent selection for productivity in a population derived from elite germplasm.
   a. 98 $S_{1:2}$ families from selected crosses of the sixth cycle (C_6) of recurrent selection were grown for seed increase during the 1994 growing season at Lewiston. A single S_3 plant from each cross was grown in the greenhouse in the winter of 1994-95 and the $S_{3:4}$ progenies planted at Lewiston for seed increase prior to testing.
   b. Residual genetic variance for yield, pod, and seed characters was measured by testing four S_0- and S_1-derived families from each of 80 C_5 crosses for a second year at Lewiston and Rocky Mount. This study will partition the observed variation into components due to C_5 parents, C_5 crosses, S_0 plants within crosses, and S_1 plants within S_0's.

6. Recurrent selection for productivity in a population derived from an interspecific cross (*A. hypogaea* / *A. cardenasii*). 36 $S_{0:1}$ families from the fifth cycle (C_5) of recurrent selection were grown in replicated trials at Lewiston and Rocky Mount. Yield was the criterion for selection, but market grades were also evaluated to identify families with potential for further selection in the cultivar development stream. The ten highest-yielding selections were planted in the greenhouse in 1995 to be crossed in diallel fashion.

B. Plant Pathology

1. Cercospora leafspot diseases: Studies were conducted to elucidate the role of climatic factors on expression of levels of partial resistance on seven North Carolina genotypes and six genotypes selected at ICRISAT in West Africa (Phytopathology 84: 1037-1043). Several genotypes were identified in this

study which expressed stable levels of resistance to *C. arachidicola* across all temperature regimes. Significant pathogen isolate by genotype interaction were also previously found for some components of resistance when genotypes were inoculated with isolates of *C. arachidicola* from various locations around the world. This study was conducted to evaluate the effect of genotype (same genotypes and temperature regimes as reported in the previous, published study) on conidial germination percentage as an additional aspect of genotype performance in diverse geographic regions of the world. Conidia applied to leaf surfaces germinated at all temperature regimes, but percentage germination differed significantly with respect to time and genotype in each regime. Fewer conidia germinated at all temperature regimes on resistant compared to susceptible genotypes, with conidia on the resistant genotype 91 PA 150 (a virginia-type derived from a cross of *A. hypogaea* and *A. cardenasii*) differing most strikingly from the others. Generally, conidia germinated more quickly and more conidia germinated overall on the susceptible NC 7 than on the other genotypes tested. A greater percentage of conidia germinated at the coolest temperature regime of 26C day / 20C night and constant 24C than in warmer temperature regimes.

2. Effects of tillage on Sclerotinia blight, Sclerotium wilt, and Cylindrocladium black root rot: Microplot studies were continued to determine the effects of minimum tillage on disease incidence of CBR, Sclerotinia blight and southern stem rot of peanut in the N.C. production region. Reduced tillage in this system was simulated by adding wheat straw to the soil surface. Maintaining organic material at the soil surface, as in reduced tillage, alters soil temperature and moisture levels. In 1992, '93, and '94, the influence of surface plant debris on disease incidence was evaluated in microplots. In 1992 soil in microplots was infested with either *Cylindrocladium parasiticum*, *Sclerotinia minor*, or *Sclerotium rolfsii* and plots were planted with NC 7, a susceptible cultivar, and NC 10C, a moderately resistant cultivar. Wheat straw was applied to selected microplots, providing 80-90% soil surface coverage. Disease incidence data were collected biweekly in 1992 and weekly in 93-94. Debris had minimal effect on incidence of southern stem rot in all years. However, soilborne inoculum density (sclerotia) was higher at the conclusion of the study (1994) in plots receiving wheat straw amendments. Time of onset and severity of CBR was enhanced by wheat straw amendments in the first year of this study (1992). In the same plots in 1993, however, disease enhancement with straw amendments was only observed on the more resistant cultivar (NC 10C). In continuous cropping Sclerotinia blight incidence was consistently suppressed by the addition of wheat straw for both cultivars in all years.

3. Integrated management of Sclerotium stem rot and Sclerotinia blight: Evaluation of management and control strategies for Sclerotinia blight by modification (genetic, mechanical pruning) of peanut canopy characteristics alone, or in combination with fungicide application, was continued in grower's fields. In addition to collecting data on disease progress, pod yield and yield-quality factors were determined for genotypes, genotype x pruning, and genotype x pruning x fungicide application. Microclimate effects resulting from canopy alterations were also studied using morphologically dissimilar genotypes (NC 7, VA 93B, NC Ac 18016, and Tamspan) and mechanical pruning (using a tractor-mounted bush-hog mower). Results indicate that pruning was as effective as fluazinam in reducing the disease in only three genotypes (NC 7, VA 93B, and Tamspan), and pruning measurable affects soil temperature and soil moisture. The combination of pruning and fluazinam did not reduce yields when compared to the highest yielding plots (fluazinam sprayed plots); however, pruning alone significantly reduced yield. An extra treatment was added this year which consisted of pruning certain plots twice during the growing season. This new treatment did not offer better control of Sclerotinia blight than the single pruning treatment, nor did pod yield decline when compared to single pruning treatments. Data was collected by counting number of "hits" (active lesions). Hull-scrape pod maturity tests (using several currently used North Carolina cultivars) revealed that pruning did not alter maturity of virginia-type peanut. A separate field trial was again conducted to determine whether plant debris left from pruning would influence the incidence of S. rolfsii severity of in the field. Our results indicated, that again, pruning did not increase the incidence of severity of *S. rolfsii*.

4. Biology of Cylindrocladium black rot resistance: Investigations are continuing on the role of various root growth parameters in Cylindrocladium black rot in peanut. Previous studies of root systems

grown in both field and greenhouse situations have shown that resistant genotypes produce less root length than susceptible genotypes, thus leading to fewer root-propagule contacts with the more resistant genotypes. Recent studies have explored the ability of genotypes to recover from artificial wounding, a treatment which is meant to mimic root pruning by the pathogen. Plants were allowed to grow for 4 weeks from time of planting, after which a portion of the soil profile was cut away and removed. Fresh soil was then replaced and the plants were allowed to grow for future sampling at either 6, 8, or 10 weeks from time of planting. Data collected from these experiments have been analyzed only superficially, but promising results already have been found. Overall, the more resistant genotypes produced more total root length than the susceptible genotypes. Although this seems to contradict findings of previous studies, these earlier studies did not include time intervals as late as those in the current wounding studies, nor did the earlier studies directly address the regrowth of roots into different localities of the soil profile as a result of wounding in another portion of the profile. Also, significant genotype by section interactions were detected, but further detailed analysis is needed to discover where these differences lie. However, when root growth into the fresh soil was analyzed separately, root length was greater for the two resistant genotypes. This preliminary data indicates that resistant genotypes are more capable of compensating for root pruning by the pathogen.

5. Pod rot research: Management and control of pod rotting pathogens of peanut in North Carolina has traditionally depended on a combination of cultural practices, host resistance, and soil-applied pesticides. Recent studies of conservation tillage practices for peanut, however, have indicated that certain soilborne pathogens are enhanced in activity by modest changes in soil temperature and moisture in no-till soils. A multiyear study was initiated to determine whether grower observations suggesting a resurgence of pod rot diseases in peanut was occurring state-wide, and the possibility that changes in tillage practices may alter the biology of soil biota. A preliminary survey was made of selected farms in Fall, 1994, to identify occurrence of specific pod rot pathogens. Both *Pythium* and *Rhizoctonia* spp. were commonly isolated; more pods, however, were infected by Pythium spp. than by *Rhizoctonia* spp. *Cylindrocladium parasiticum*, *Sclerotinia minor*, and *Sclerotium rolfsii* were also identified from individual fields. Microplot tests were established to investigate the influence of winter covercrops (wheat, rye, oats) in conjunction with tillage (no-till, burial of debris) to determine effects of cultural practices on *Pythium*, *Rhizoctonia* and a mixture of the two as factors in subsequent years pod rot incidence.

6. Biology and control of seed transmission of CBR: Field seed transmission studies have been conducted during the last three years. The purpose of these tests were twofold. The first goal was to determine if symptomatic peanut seed transmit the CBR fungus and cause diseased plants. The second goal was dependent on the first. If seed transmission occurred, seed treatments were to be tested to determine if they eliminated or lowered the number of diseased plants. All symptomatic seed used for seed transmission studies and for greenhouse fungicide screenings were removed from commercial seed bins awaiting the application of seed treatments. These seed have yielded infected plants from untreated seed during three seasons (1992, 1993, 1994) and from seed treated with Vitavax PC in 1993. The differences experienced during these three years indicate this pathogen is environmentally sensitive. Two different approaches, fungicidal seed treatments and electronic sorting, are being researched with funds provided and progress is being made to control seedborne CBR. Methodology for screening fungicidal seed treatments is progressing. The rate of seed transmission is variable among seed lots and among years. Since the rate is often low in commercial seed lots, very large numbers of seeds are required to screen for a given fungicide. In initial studies, the seed transmission rate of the commercially conditioned seed used was very low and isolation methods were necessary to detect infection. A high rate of transmission is needed to adequately screen and compare treatments. Different methods are being investigated to circumvent the low transmission rate problem. Work in Virginia indicated the pathogen can be in the seed coat and in the cotyledons. We are therefore artificially inoculating the seeds in these locations. Preliminary experiments inoculating the seed coats indicate this method is a viable alternative.

7. Selection and evaluation procedure for genotypes containing multiple pathogen resistances: Preliminary studies have indicated that germplasm lines such as NC 3033 and NC Ac 18016, which possess high levels of resistance to Cylindrocladium black rot (CBR) caused by *Cylindrocladium parasiticum*, also have resistance to Southern stem rot caused by *Sclerotium rolfsii* and Sclerotinia blight caused by *Sclerotinia minor*. Two disease nurseries were established at the Upper Coastal Plain Research Station in order to evaluate resistance to *S. minor* and *S. rolfsii* in breeding lines of virginia-type peanut that were selected primarily for resistance to CBR. Sixteen genotypes were planted in four, two-row plots in each nursery and were inoculated in August with oat grains infested with the appropriate pathogen. Percentage disease incidence and percentage mortality were determined from counts taken at digging. Lesion numbers also were counted on selected plants. Genotypes differed significantly in resistance to Sclerotinia blight and stem rot in 1993 and 1994. In both years, incidence of Sclerotinia blight was least on Chico, a field-resistant Spanish type. NC 3033 had slightly, but not significantly, more disease than Chico. Incidence of Sclerotinia blight on three of the virginia-type breeding lines did not differ significantly from incidence on NC 3033 in 1993; two of the lines had levels of disease equal to that on Chico. In 1994, disease incidence on four virginia-type breeding lines did not differ from that on NC 3033. Breeding lines N92054C and N92056C, which originated from a cross of NC-V 11 and NC Ac 18016, had superior resistance to Sclerotinia blight in both years. Resistance to Southern stem rot also was found in these lines in both years.

C. Molecular biology. During the current reporting period, we have made significant progress toward the development of transgenic peanut lines with enhanced resistance to peanut stripe virus (PStV). We have also improved the technology of peanut transformation by decreasing the time required to produce transgenic cell lines, and by improving the fertility of transgenic plants.

1. Transformation experiments. Transforming DNA consisting of a vector (plasmid) carrying the full-length coding sequence for the coat protein gene of PStV, and a chimeric gene encoding the selectable marker hygromycin phosphotransferase (HPT) was delivered to embryogenic cultures derived from peanut cultivars Toalson and NC 7. DNA transfer was carried out by microprojectile bombardment. Bombardment and hygromycin selection were essentially according to published protocols, except that cultures were selected on antibiotic for shorter periods than were used previously. Culture protocols were also altered, in some instances, by eliminating the liquid selection step which was used previously. This modification was found to enhance transformation efficiency by as much as two-fold, but did not appear to alter the frequency of escapes (i.e., non-transformed plants not killed by antibiotic) which were recovered.

   Using these altered protocols, ten transgenic lines derived from cv. Toalson were recovered. From each line, numerous cuttings were taken and rooted in a mist bench. These rooted cuttings represented a genetically uniform population of plants with which a replicated efficacy trial could be established. This procedure not only produced genetically identical individuals, but also allowed testing earlier than would have been possible if tests had been carried out using seedlings.

2. Efficacy testing. Rooted cuttings were inoculated with PStV by mechanical inoculation, under containment, in a temperature-controlled greenhouse. Of this first group of ten transgenic events, two produced plants which showed reduced and/or delayed symptom formation, compared with untransformed controls. Further, some additional lines appear to lose symptoms after an initial infection period. These lines have now been tested in two separate replicated trials, and resistance appears to be stable. Further, all of these lines have now set pods, and some R_1 progeny have been recovered.

   Resistant lines are now undergoing complete molecular and biochemical characterization. Although reported results of resistance against potyviruses in other species suggests that resistance is independent of coat protein accumulation, Western blot analysis of resistant lines show the presence of detectable amount of coat protein. This analysis will be repeated to confirm this correlation. Southern blot analysis is also being carried out to determine the relationship between transgene copy number and

virus resistance. Northern (RNA) blot analysis will also be used to measure the amount of transcript produced in resistant and susceptible transgenic plants.

3. Finally, numerous additional transformed lines have been recovered from transgenic cell lines of both Toalson and NC 7. These lines have all been transferred to the greenhouse. Efficacy testing will begin on these materials as soon as they can be sampled without compromising fertility of the primary transformants.

D. Cytogenetics

1. Evaluating germplasm for disease and insect resistances
   a. Twenty backcross progenies lines derived from NC 5 or NC 6 crosses with advanced generation interspecific hybrids between *A. hypogaea* and *A. cardenasii* were evaluated in replicated field plots for resistance to *Cercospora arachidicola*. High levels of resistance were observed in several of the lines. Although immunity was not observed, the resistance levels were higher than any other lines observed by the PI. Several of the lines also have large seeds. Thirteen of these lines were planted in a replicated sprayed and non-sprayed test in the field and independently evaluated for *C. arachidicola* resistance and yield. Most entries did not vary greatly between sprayed and non-sprayed plots. Although disease resistance was high, yields were low as compared to the cultivars produced in the region. Seeds were harvested from single plants of the agronomically most promising lines were grown in a non-replicated experiment.
   b. To increase yields of the leafspot-resistant lines, a second backcross was made to the large-seeded cultivars NC 6, NC 7 and NC 9. F_2 progenies from 55 crosses were planted in the field from which more than 300 single plant selections were made the last week of September based on leafspot resistance. An additional 70 lines in the F_3 generation plus 75 lines were in the F_4 generation were evaluated for leafspot resistance and selections made from the materials. Seed traits were recorded and 197 lines selected with acceptable size and shape.
   c. Interspecific hybrids derived from *A. cardenasii*, *A. diogoi*, *A. batizocoi*, *A. correntina*, and *A. stenosperma* were evaluated in the field for resistance to thrips (*Frankliniella fusca*), corn earworm (*Helicoverpa zea*), potato leafhopper (*Empoasca fabae*) and southern corn rootworm (*Diabrotica undecimpunctata*) in replicated field experiments. One hundred seven hexaploid lines, 30 lines in early generations after backcrossing hexaploids to *A. hypogaea* to form pentaploid progenies, and 46 late generation lines at the tetraploid level were evaluated in replicated experiments. Little resistance to thrips was found whereas high levels of resistance to corn earworm and leafhopper were observed. The numbers of southern corn rootworms in the field were too low for reliable ratings. Twelve lines were observed morphologically to record descriptor information to be used for germplasm registration.
   d. Eighty four backcross progenies in the F_5 generation were evaluated for resistance to an insect complex in replicated field tests and compared with three cultivated genotypes plus three resistant *A. hypogaea* x *A. cardenasii* lines. High levels of resistance were observed for leafhopper and corn earworm. Southern corn rootworm damage was of insufficient intensity to rate in the field, and no resistance to thrips was observed.

2. Introgressing germplasm from wild *Arachis* species to *A. hypogaea*
   a. Forty-six introgression lines from a cross between *A. hypogaea* and *A. cardenasii* were analyzed for the introduction of *A. cardenasii* chromosome segments. Seventy RAPD primers, expressing from one to four *A. cardenasii* specific bands were used to evaluate the set of introgression lines. Introgressed segments were detected by many RAPD markers and was confirmed by using RFLP markers. Ten of 11 linkage groups were found with *A. cardenasii* introgression. Several of the markers were associated with disease or insect resistances.

      Triploid interspecific hybrids were produced by crossing the two species *A. cardenasii* and *A. batizocoi* with the two *A. hypogaea* cultivars NC 6 and Argentine. Fertility was restored at the hexaploid level in the four hybrids after colchicine treating vegetative meristems. Hexaploids

were self-pollinated for four generations (C_1, C_2, C_3 and C_4) and backcross families derived from each generation by crossing with the respective *A. hypogaea* parent. RAPD species-specific markers of know map location were utilized in this study. Wild species germplasm retention decreased with every generation of selfing after backcrossing (within a cycle), probably due to preferential loss of wild species chromosomes. This effect was more pronounced in cv. Argentine and in *A. batizocoi* crosses. A decay in wild species retention was also observed for every cycle in BC_1F_2 and BC_1F_3 families, although the reason for this is unclear. Introgression analysis of 40- and near 40-chromosome derivatives from the four crosses confirmed previous reports that reciprocal recombination and/or translocations are the predominant mechanisms of introgression. No evidence was found for preferential introgression into specific linkage groups.

Two dominant genes conditioning resistance to the root-knot nematode *Meloidogyne arenaria* were identified in a segregating F_2 population derived from the cross of 4x (*A. hypogaea* x *A. cardenasii*)-GA 6a and PI 261942. One RAPD marker was linked to both the gene conditioning galling and to another gene conditioning egg number. The marker was mapped to linkage group 1 on the RAPD map. This fragment was cloned and used to generate a pair of primers that specifically amplified this locus and an RFLP probe. Neither of the two genes was found in other introgression lines or elite breeding materials. This represents the first molecular markers linked with a resistance gene in peanut and the first report of two physiological responses to nematode attack associated with two genetic factors.

b. One hundred seven hexaploid and thirty pentaploid lines in advanced generations were propagated in field for seed increases. In addition, hexaploids and pentaploids from the introgression study with *A. batizocoi* and *A. cardenasii* were propagated for seed increases. Hexaploids were hybridized with NC 6 and Argentine to obtain additional pentaploid progenies.

II. Philippines

A. Development of a Philippine Seedboard Variety in 1994. One Philippine seedboard variety, PSB Pn 2, was released in 1994. PSB Pn 2 is locally known as UPL Pn 12 or Biyaya 12. It is a valencia peanut variety derived from the three-way cross between (UPL Pn 2 x FESR 1) x NC 6. Its seed yield is higher than both check varieties. It exceeded the seed yield of BPI Pn 2 by 18% and PSB Pn 1 (released in 1992) by 13% in 12 tests in the national cooperative yield trials during two dry season croppings across seven locations. It has better resistance to peanut rust and late leafspot diseases than both check varieties. Similar to PSB Pn 1, it has resistance to leafhopper damage. It has moderate resistance to *Aspergillus flavus* invasion using dry seed resistance test, and it has high seed viability even after 6 months pod storage at ambient storage condition. It has two to four seeds per pod, and the majority of the pods are three-seeded. It has likewise a tan seed coat. Similar to UPL Pn 4, it has a sweet taste and is recommended to be utilized for boiling, peanut brittle, and peanut butter manufacture.

B. High Seed Viability Lines. Lines with high seed viability were identified 8 months after storage as seeds under conditions where no $CaCl_2$ was used. These include IPB Pn 87-64BC-8, IPB Pn 87-65BC-22, IPB Pn 87-66BC-23, IPB Pn 87-67BC-20, and IPB Pn 88 19-6. Likewise, PSB Pn 1 or UPL Pn 10 had comparable seed viability potential.

C. Selection and Inclusion of 13 New Entries in the National Cooperative Tests. A total of five new entries were included in the national cooperative tests across nine locations in the Philippines in 1995. These new entries included IPB Pn 88 19-6, IPB Pn 87 24-28, IPB Pn 88 19-45, IPB Pn87-65BC11, and IPB Pn87-66BC13.

III. Thailand

A. Thailand Department of Agriculture. The peanut breeding work of the TDOA aims to develop cultivars with high yield, early maturity, resistance to rust, leafspots and aflatoxin, and to develop large-seeded peanut.

Activities in 1994 were concentrated on yield testing of various groups of materials at different evaluation stages. Breeding lines selected for the various breeding objectives were initially tested in the preliminary yield trials. At the preliminary yield trial stage, the lines were grouped according to the individual breeding objectives and tested separately. At the standard, regional, and farm trial stages, the lines were merged into three series of trials: a medium-seeded type, large-seeded type, and early maturity lines. Promising lines from each trial were selected for entry into the next stage of testing in order to evaluate their adaptation and stability in diverse environments. All trials except the farm trials were conducted at the research centers and experiment stations of the Field Crops research Institute. Tests were conducted under rain-fed conditions in the rainy season and under irrigation in the dry season.

1. Preliminary Yield Trials.
   a. Breeding for high yield. The main objective of this program is to improve yield potential. In 1994, there were four sets of Preliminary Yield Trials. The first and second sets were the coordinated trials conducted with Khon Kaen University. The first set was conducted at the Khon Kaen Field Crops Research Center in the dry season; the second set was conducted at Roi-et Field Crops Experiment Station during the rainy season. Breeding line (ICGS 20 / ICG 2337 NC Ac 02956)-1 was the top-yielding line in the first yield trial (2025 kg ha^{-1}). (ICG 2375 NC Ac 02938 / ICGS 4)-2 gave the highest yield in the second trial (2037 kg ha^{-1}). The third and fourth sets consisted of 27 high yielding lines from Khon Kaen Field Crops research Center and three checks. The third trial was conducted at Kalasin Field Crops Experiment Station in the dry season; the fourth at the Mahasarakham Field Crops Experiment Station in the rainy season. The top yielding lines in the two trials were (KK4 / NC 8C)-40-9-28, (KK 60-3 / (G37 / PI 259747))-36-8-20, (KK4 / NC 8C)-4-3-10, and (Tainan 9 / Non-Nodulating)-4-8-2.
   b. Breeding for early maturity. Entries in a preliminary yield trial were lines selected from ICRISAT materials. The trial was conducted at Kalasin Field Crops Experiment Station in the rainy season. Yields in this trial were low because of drought at the flowering and pegging stages. A check variety, Tainan 9, gave the highest average yield (968 kg ha^{-1}).
   c. Breeding for resistance to aflatoxin. A preliminary trial was conducted in the dry and rainy seasons at Kalasin and Khon Kaen. Seventeen lines from ICRISAT and seven checks were evaluated. A check variety, Khon Kaen 4, and the breeding line VRR 245 gave the highest average yields (1381 and 1359 kg ha^{-1}, respectively). The high yielding lines were selected for further testing of resistance to *A. flavus*.
   d. Breeding for large-seeded type. Entries in the preliminary yield trial were 33 lines and a check variety, Khon Kaen 60-3 (NC 7). The trial was conducted in the rainy and dry seasons at Khon Kaen and Roi-et. The highest yielding lines were (KK 60-3 / Non-Nodulating)-91, (KK 60-3 / Non-Nodulating)-13, (KK 60-3 / Non-Nodulating)-9, and ICGV 88362.

2. Peanut Standard Yield Trial. Entries in the standard yield trials are promising lines identified in preliminary yield trials. In 1994, four standard yield trials were conducted. The first and second standard yield trials were the coordinated trials with Khon Kaen University. The first trial was conducted in the dry season at Phitsanulok, Kalasin, and Sakon-Nakorn. (Tainan 9 / KK 60-3)-3-9-18 and check variety Khon Kaen 60-1 were the top-yielding entries. Five lines were selected from this trial for evaluation in the rainy season trial of 1994. The second trial was conducted in the dry and rainy seasons at Khon Kaen, Phitsanulok, Kalasin, Chiang Mai, Rayong, Roi-et, Loei, Mahasarakham, and Khon Kaen University. The highest yielding lines were ((MGS 9 /

Chico)-12-16-5 / KK60-3)-117, ((MGS 9 / Chico)-12-16-1 / KK60-3)-20, (ICG 2956 SM5 / RCM 387)-3, and (KK60-1 / ICG 404SB NC Ac 17090)-2.

The third trial included 12 early maturing lines and two checks and was conducted at 11 locations. A check variety, (MGS 9 / Chico)-12-16-5 gave the highest average yield (1462 kg ha^{-1}). The entries in the fourth standard yield trial were 13 large-seeded lines and a check, Khon Kaen 60-3. This trial was conducted in the dry and rainy seasons at Chiang Mai, Khon Kaen, Nakornsawan, Praputtabhat, Petchaboon, and Mahasarakham. (KK60-3 / Tainan 9)-23 gave the highest average yield (1480 kg ha^{-1}).

3. Peanut Regional Yield trials. Superior entries in the standard yield trial were selected for inclusion in the regional yield trial which is conducted at more locations. There were four regional yield trials. The first and second sets were coordinated yield trials. Eleven entries in the first trial were evaluated at Phitsanulok and Kalasin in the dry season. Check variety Khon Kaen 60-1 was the highest yielding line (1850 kg ha^{-1}). In the second trial, seven lines were tested in the rainy season at Rayong, Ubon, Mahasarakham, and Khon Kaen University. ICGS 110 was the best line in this trial. Entries in the third trial were nine high-yielding lines and three checks. The trial was conducted in the dry and rainy seasons at Chiang Mai, Khon Kaen, Kalasin, Rayong, Roi-et, and Srisamrong. (Chico / UPL Pn4)-8-16-12 was the top yielding line. Four lines from this trial were selected for further evaluation in the Farm Trial in the dry season of 1995.

4. Peanut Varietal Farm Trial. Two sets of promising entries selected from the Regional Yield Trials were entered in the Farm Trial conducted in farmers' fields in 1994. The first set was selected from the high-yielding group. It included five breeding lines and two check varieties, Tainan 9 and Khon Kaen 60-1. (Tainan 9 / RCM 387)-12-3-11 gave the highest average yield and had larger average seed size than the other lines and checks. The second set of entries included four entries from the large-seeded group and two checks, Khon Kaen 60-1 and Khon Kaen 60-3. This trial was conducted in farmers' fields at Chiang Mai, Nakornsawan, Praputtabhat, Petchaboon, Khon Kaen, and Rayong in the rainy season of 1994. None of the test lines performed better than the Khon Kaen 60-3.

B. Khon Kaen University. The main objectives of the KKU breeding program are resistance to rust and leafspot, early maturity, and adaptation to growth before or after rice without irrigation. Other objectives include resistance to insects, improvement of the large-seeded and boiling types, and increased biological nitrogen fixation. Work completed in 1994 included evaluation of yield of lines selected from different subprograms. Promising lines were selected from each set of breeding materials and submitted to the TDOA for multiple-site testing.

1. Breeding for after-rice unirrigated condition. Five yield trials were conducted in the 1993-94 after-rice season. These included a preliminary yield trial, an intermediate yield trial, an elite yield trial, an advanced yield trial, and an on-farm trial. The test entries, more than 80 lines, were selected from the 1992-93 after-rice trials. All trials were conducted during the dry season in paddy fields with high residual soil moisture. The on-farm trial was conducted at three sites: Khon Kaen, Surin, and Korat. All other trials were conducted at Khon Kaen. In the on-farm trial, ICGS 11 gave the highest yield among the test entries. Khon Kaen 60-1 gave higher yield in the advanced yield trial while Tainan 9 gave the highest yield in the elite yield trial. (Lonyun 6101 / (X52 X-X-3X-BX / Chico)-8-1-2)-F_6-30 performed better than check variety Khon Kaen 60-1 in the intermediate yield trial. (TMV 3 / ((X52 X-X-3X-BX / Chico)-8-1-2)-F_6-12 was the best line in the preliminary yield trial.

2. Breeding for before-rice growing conditions. Six yield trials, including two preliminary yield trials, an intermediate yield trial, two advanced yield trials, and an on-farm trial, were conducted using 110 lines selected from the 1993 trials. The on-farm test was conducted at three sites; all tested entries performed better than check varieties Tainan 9 and Khon Kaen 60-1. ICGS(E) 173 and (MSG 9 / Robut 33-1)-5-2-2-4 gave the highest yields in this trial. All other trials were

conducted at Khon Kaen University. (ICGS / ICGS 404 SM NC Ac 17090) and (ICGS 2596 SM-5 / RCM 387)-F_7-2 gave the highest yields in the two advanced tests. (ICGS 2375 NC Ac 2938 / ICGS 4)-F7-1 was among the top entries in the intermediate yield trial. (Lampang / (MGS-9 / Robut 33-1)-5-2-2-4)-F_6-13 and ((X5-2-X-X-X-3X / Chico)-8-1-2 / (MGS-9 / Robut 33-1)-5-2-2-4))-F_6-2 yielded 51% and 61% more than the check Khon Kaen 60-1 in the two preliminary yield trials.

3. Breeding for high yield. Three yield trials were conducted under this breeding objective, including a preliminary yield trial, an intermediate trial, and an advanced trial. More than 50 lines were selected form the 1993 rainy season trials for entry in this series of tests. (UPL Pn4 / Lampang)-F_6-41 was the top yielding entry in the advanced test, (Moket / NC 7)-F_4-41 was the best line in the intermediate trial, and ((MGS-9 / Chico)-16-1-2 / PLPS 12)-F_4-1-2 was the best in the preliminary trial.

4. Breeding for earliness. Preliminary and intermediate trials of a total of 15 lines were conducted in the 1994 rainy season. ICGV 87003 gave the highest yield in the intermediate trial while (ICGS(E) 229 / JL 24)-F_5-3 outyielded the performance check, Tainan 9, in the preliminary trial.

5. Improvement of large-seeded type peanut. Two preliminary yield trials of large-seeded materials were conducted in the 1994 rainy season at Khon Kaen University. (NC Ac 17090 / B_1)-10-B1 and RCM 387 were the top-yielding entries in the two trials, but both have smaller seeds than Khon Kaen 60-3, the large-seeded check.

6. One preliminary yield trial of foliar disease-resistant lines was conducted at Khon Kaen University in the 1994 rainy season. (GP-NC 343 / Tainan 9)-F_7-8 was the top-yielding line among 15 test entries.

TRAINING

Surname	Sex	Univ.	Department	Degree	Date degree received	CRSP support
Garcia (Argentina)	M	NCSU	Crop Science	PhD	August, 1995	Total
Menon (India)	F	NCSU	Crop Science	PhD	--	Total
Utomo (Indonesia)	M	NCSU	Crop Science	PhD	--	Partial
Salas (Peru)	M	NCSU	Crop Science	PhD	--	Partial

R.M. Abilay traveled to Tulsa, Oklahoma, on July 13-18, 1994, to attend the 1994 meeting of the American Peanut Research and Education Society and to meet with Dr. T.G. Isleib and other peanut collaborators regarding the CRSP breeding project in the Philippines.

Dr. Abilay, Dr. Aran Patanothai, and Dr. Sanun Jogloy traveled to Rosslyn, Virginia, to participate the Peanut CRSP Symposium, Impacts and Scientific Advances through Collaborative research on Peanut, March 29-31, 1995.

PUBLICATIONS AND PRESENTATIONS

Abilay, R.M., and L.A. Lantican. 1995. PSB Pn 2 (UPL Pn 12) - A newly released valencia peanut variety at the Institute of Plant Breeding, University of the Philippines at Los Baños. Phil. J. Crop Sci. 20(Suppl. No. 1): 9.

Abilay, R.M., and E.E. del Rosario. 1995. Promising peanut lines with high seed viability. Phil. J. Crop Sci. 20(Suppl. No. 1): 9.

Barrientos, L., T.G. Isleib, and H.T. Stalker. Combining abilities of lines derived from an interspecific cross, *Arachis hypogaea / A. cardenasii*. (Abstr.) Proc. Am. Peanut Res. Educ. Soc., Inc. 26:32.

Bianchi-Hall, C.M., R.D. Keys, and H.T. Stalker. 1994. A note on use of seed protein markers for identification of aflatoxin resistance in peanut. Peanut Sci. 21: 159-161.

Bianchi-Hall, C.M., R.D. Keys, and H.T. Stalker. 1994. Use of protein profiles to characterize peanut cultivars. Peanut Sci. 152-159.

Clemente, T.E., A.K. Weissinger, M.K. Beute, M.C. Black, and H.T. Stalker. n.d. Identification of *Arachis* germplasm with resistance to tomato spotted wilt virus. Peanut Sci. (submitted 5/93).

Dhesi, J.S., and H.T. Stalker. 1994. Enhancing techniques for studying mitotic peanut chromosomes. Peanut Sci. 21: 92-94.

Feng, Q.L., H.E. Pattee, and H.T. Stalker. 1994. In vitro reproductive development of a diploid wild species *Arachis* duranensis. Peanut Sci. 139-143.

Feng, Q.L., H.T. Stalker, and H.E. Pattee. n.d. Peanut plant regeneration through *in vitro* culture of peg tips of *Arachis hypogaea*. Crop Sci. (accepted).

Garcia, G.M., H.T. Stalker, and G.A. Kochert. 1995. Introgression analysis of an interspecific hybrid population in peanuts (*Arachis hypogaea* L.) using RFLP and RAPD markers. Genome 38: 166-176.

Lantican, L.A., and R. M. Abilay. 1995. Performance of 49 peanut entries under reduced and full sunlight conditions. Phil. J. Crop Sci. 20(Suppl. No. 1): 8.

Shew, B.B., M.K. Beute, and H.T. Stalker. 1994. Towards sustainable peanut production: Progress in breeding for foliar and soil-borne pathogens of peanut. Plant Dis. 78: 1259-1261.

Stalker, H.T, G.M. Garcia, B.B. Shew, M.K. Beute, T.G. Isleib, and G. Kochert. Introgression from *Arachis cardenasii* to *A. hypogaea*. (Abstr.) Proc. Am. Peanut Res. Educ. Soc., Inc. 26:33.

Stalker, H.T., J.S. Dhesi, and G.A. Kochert. 1995. Genetic diversity within the species *Arachis duranensis* Krapov. & W.C. Gregory, a possible progenitor of the cultivated peanut. Genome 38: 1201-1212.

Stalker, H.T., M.L. Nickum, J.C. Wynne, G.H. Elkan, and T.J. Schneeweis. 1994. Evaluation of *Arachis* species for biological nitrogen fixation capacity. Peanut Sci. 21: 55-60.

Tallury, S.P., H.T. Stalker, and H.E. Pattee. 1995. Early reproductive ontogeny in interspecific crosses of *A. hypogaea* and section Arachis species. Ann. Bot. 76: 397-404.

Utomo, S.D., A.K. Weissinger, H.T. Stalker, and T.G. Isleib. n.d. High efficiency of peanut regeneration using a nonimbibed immature leaflet culture method. Peanut Sci. (in review).

Waliyar, F., B.B. Shew, H.T. Stalker, T.G. Isleib, R. Sidahmed, and M.K. Beute. 1994. Effect of temperature on stability of components of resistance to *Cercospora arachidicola* in peanut. Phytopathology 84: 1037-1043.

Waliyar, F. B.B. Shew, H.T. Stalker, T.G. Isleib, R. Sidahmed, and M.K. Beute. Effect of temperature on stability of components of resistance to *Cercospora arachidicola* in peanut. (Abstr.) Proc. Am. Peanut Res. Educ. Soc., Inc. 26:47.

Presentations

Abilay, R.M., and T.G. Isleib. 1995. Breeding for large peanut seeds in the Philippines. Paper presented at the P-CRSP International Research Symposium and Workshop on Impacts and Scientific Advances Through Collaborative Research on Peanut, 29-31 March. Washington, DC.

Barrientos, L., T.G. Isleib, and H.T. Stalker. Combining abilities of lines derived from an interspecific cross, *Arachis hypogaea / A. cardenasii*. Am. Peanut Res. Educ. Soc., Inc., Tulsa, OK, July 12-15, 1994.

Isleib, T.G., and O.D. Smith. Stalker, H.T, G.M. Garcia, B.B. Shew, M.K. Beute, T.G. Isleib, and G. Kochert. Introgression from *Arachis cardenasii* to *A. hypogaea*. Am. Peanut Res. Educ. Soc., Inc., Tulsa, OK, July 12-15, 1994.

Waliyar, F. B.B. Shew, H.T. Stalker, T.G. Isleib, R. Sidahmed, and M.K. Beute. Effect of temperature on stability of components of resistance to *Cercospora arachidicola* in peanut. Am. Peanut Res. Educ. Soc., Inc., Tulsa, OK, July 12-15, 1994.

Shew, B.B., M.K. Beute, and H.T. Stalker. Towards sustainable peanut production: progress in breeding for resistance to foliar and soil-borne pathogens of peanut. Amer. Phytopath. Soc., Albuquerque, NM.

Releases

Release of CBR-resistant NCSU breeding line N90013E as 'NC 12C', pending approval by institutional committees and administration.

Release of UPLB breeding line IPB Pn 85 2-40 in the Philippines.

PLANS FOR FY 1995-96

I. Research thrusts / endeavors planned

A. U.S.

Breeding

1. Select intensively for large-seeded adapted lines with resistance to Cylindrocladium black rot (CBR) caused by *Cylindrocladium parasiticum*.
   a. Six lines with superior agronomic performance and resistance to CBR will be crossed in a diallel mating along with four agronomically lines exhibiting resistance to early leaf spot. F_1 seeds will be sent to the winter nursery at the USDA Tropical Agriculture Research Station at Mayaguez, Puerto Rico.
   b. F_2 populations from a partial factorial mating of three sources of resistance to CBR with four breeding lines possessing superior seed attributes and high yield will be screened for resistance in a naturally infested field in Bertie County, NC. Selected $F_{2:3}$ families will be sent for increase to the winter nursery. $F_{2:4}$ families will be planted in the infested area in 1996.
   c. Selection for resistance and pod attributes will be practiced on infested soil among and within $F_{2:4}$ families derived from 68 $_2$F plants selected during the 1993 growing season. F progenies will be grown in a replicated test on infested soil in 1996.
   d. 96 $F_{4:5}$ families and checks will be evaluated on infested soil for CBR incidence in a completely random design with unequal replication to allow for variable seed number. These

families will be increased on uninfested soil at the NCDA Peanut Belt Research Station at Lewiston, NC, and resistant families will be screened for pod and seed characters.

e. 57 F_4- and F_6-derived families selected will be compared on infested soil with checks for CBR resistance in a replicated test and for yield and market grade at Lewiston. Four of the lines were first evaluated in the CBR Advanced Test in 1992; three in 1993 and seven in 1994. Five will be included in the Advanced Yield Test leading to cultivar release. Also included in the test are six lines derived from a cross between CBR-susceptible cultivars NC 7 and NC 9. These six were found to exhibit partial resistance to CBR.

2. Selection for resistance to early leafspot (*Cercospora arachidicola*).
   a. 17 F_4- and F_5-derived families from a program of selection for resistance to early leafspot will be evaluated for defoliation, yield, and market grades in replicated tests at Lewiston in the 1995 growing season.
   b. 86 F_2 populations derived from crosses between NCSU selections with resistance to early leaf spot and University of Florida selections resistant to late leaf spot (*Phaeoisariopsis personata*) will be grown without chemical control of leafspot to permit selection for resistance within populations.
   c. 85 $F_{3:4}$ families derived from 69 leafspot-resistant F_2 plants (selected on the basis of defoliation) will be grown without chemical control of leafspot to permit selection for resistance among and within families.
   d. 184 $F_{4:5}$ families derived from at least one leaf spot-resistant parent but not previously subjected to the disease will be grown without chemical control of leafspot to permit selection for resistance among families.
   e. 61 $F_{1:5}$ families developed by single-seed descent from crosses involving at least one leafspot-resistant parent grown without chemical control of leafspot to permit selection for resistance within families.
   f. In 1994, three lines selected for resistance to early leafspot and two susceptible lines with excellent yield and grade characteristics were mated with two highly resistant tetraploid lines developed by H.T. Stalker through introgression of germplasm from *A. cardenasii* into *A. hypogaea*. F_1 plants will be back-crossed to their respective large-seeded virginia parents.

3. Selection of early-maturing large-seeded virginia lines with good storability.
   a. Populations have been developed from crosses of virginia cultivars with F435, a spanish line with an extremely high ratio of oleic to linoleic acid in the fatty acid profile of the oil. F_4-derived lines homozygous for the two recessive genes conferring the trait will be backcrossed to their respective adapted parents. F_1 hybrids will be planted in the greenhouse for backcrossing to the cultivar.
   b. 18 advanced F_5- and F_6-derived selections from crosses of NC 7 with NC 9 and Florigiant will be evaluated for performance at early (September 15) and conventional (October 1-7) digging dates.

4. Incorporation of arthropod pest resistance into *A. hypogaea*. F_4 progeny will be grown of crosses between large-seeded lines and a set of arthropod-resistant selections including tetraploid lines derived from an interspecific cross of *A. hypogaea* PI 261942-3 with *A. cardenasii* 10017. Single-plant selection will be performed and $F_{4:5}$ populations grown in 1995 as part of a program of inbred line selection.

5. Recurrent selection for productivity in a population derived from elite germplasm.98 $S_{2:4}$ families from selected crosses of the sixth cycle (C_6) of recurrent selection will be grown for seed increase 1995 growing season at Lewiston. A replicated test of the $C_6S_{2:5}$ families will be planted in 1996.

6. Recurrent selection for productivity in a population derived from an interspecific cross (*A. hypogaea* / *A. cardenasii*). The ten highest yielding selections form the 1994 trial of $S_{0:1}$ families from the fifth cycle (C_5) of recurrent selection will be intermated in a half diallel cross. F1 seeds will be sent to Puerto Rico for increase.

Plant Pathology

7. Studies will continue to examine the effects of tillage on Sclerotinia blight, Sclerotium wilt, and Cylindrocladium black root rot. Microplots will be infested with *S. rolfsii*, *S. minor*, or *C. parasiticum* in the presence or absence of surface litter (wheat straw) which may enhance or suppress disease development through several mechanisms. Two peanut genotypes will be evaluated. Soil temperature, moisture, and disease progress will be monitored in plots to determine whether surface litter enhances or inhibits diseases caused by these pathogens.

8. Peanut canopies can be modified by mechanical pruning or by selection of genotypes with the desired canopy characteristics. Canopy modification may affect development of Sclerotium wilt and Sclerotinia blight by making the microenvironment less favorable for disease development. These modifications also may allow better penetration of fungicides to target sites near the crown of the plant. Studies will be conducted to determine the effects of peanut genotype, mechanical pruning, and fungicide use on development of Sclerotinia blight. Companion studies will be used to determine if foliar pruning suppresses Sclerotium wilt by altering microenvironment or enhances disease by creating a food base for the fungus. The impact of pruning on yield will also be determined.

9. Greenhouse and field studies have indicated potential for transmission of *C. parasiticum* by peanut seed. Experiments will be conducted to determine the effects of peanut genotype, environment, and production practices on seed infection. Other studies will examine effects of inoculum source, environment, and inoculum survival on transmission rates. Management strategies to minimize occurrence of *C. parasiticum* in commercial seed trade will be developed.

10. Leaf spot resistant selections from Drs. Isleib and Stalker's selection programs will be evaluated for disease development in isolated plots.

11. Resistance of species-derived lines to *S. rolfsii* will be evaluated in field trials to confirm the results of greenhouse testing.

Molecular Biology

12. Selection and regeneration of transformed cultures and plants and efficacy testing will continue. Plants regenerated from transgenic cultures will be analysed by DNA hybridization to demonstrate the presence of the transgenes. Transgenics will then be propagated vegetatively to provide populations for efficacy testing.
    a. We will continue to conduct transformation experiments to produce stable, virus-resistant lines.
    b. Efficacy testing will consist of inoculating multiple vegetative clones of transgenics with the PStV isolate from which the coat protein gene was originally cloned. Inoculation will be carried out under appropriate containment conditions. Where necessary, transgenics will be inoculated repeatedly to insure unambiguous identification of resistant lines.
    c. Virus-resistant transgenics will be self-pollinated and backcrossed to the donor line. Self pollination of subsequent generations will be continued until materials suitable for inclusion in breeding programs are identified. Back-cross progeny will be analyzed by DNA hybridization to determine the number and linkage relationships of introduced sequences.
    d. If possible, we will test resistant lines against multiple isolates of Peanut Stripe Virus. This should allow either selection of lines resistant to a broad array of isolates, or clear delineation of the limits of the resistant genotypes when challenged with variants of the virus. Such information is critical if these materials are eventually to be deployed in multiple geographical locations, where variation of the pathogen is to be expected.

Cytogenetics

13. Evaluating germplasm for disease and insect resistances
    a. *C. arachidicola* and *P. personata* resistances. Selections from four-way hybrids between large-seeded cultivars, leafspot-resistant cultivated plant introductions, and an *A. hypogaea* / *A. cardenasii* interspecific hybrid will be evaluated in the field for resistance to *C. arachidicola*.

Disease ratings will be made in late August and in late September. Yield data will be collected for sprayed and unsprayed plots. Leaves will be collected from a sample of plants of each entry for *P. personata* evaluations in the greenhouse.

b. Selections from the population described in 1a above were backcrossed to NC 6. $F_{2:3}$ families will be grown in a progeny test of selected F_2 plants to make preliminary evaluations for resistance to *C. arachidicola*.

c. Advanced generation interspecific hybrids will be evaluated for resistance to thrips (*Frankliniella fusca*), corn earworm (*Helicoverpa zea*), potato leafhopper (*Empoasca fabae*), and southern corn rootworm (*Diabrotica undecimpunctata*) in replicated field tests. This will complete multiple-year testing for insect resistance. Lines are expected to be released from these *A. hypogaea* / *A. cardenasii* hybrids after the data is analyzed.

d. Selected progenies derived from interspecific hybrids which were backcrossed to *A. hypogaea* lines are currently in the F_5 generation. These progenies will be tested for resistance to the insect complex of North Carolina. Selection for conformation to the virginia pod and seed type will also be practiced, and superior lines will be increased for more in-depth agronomic evaluations.

14. Introgressing germplasm from wild *Arachis* species to *A. hypogaea*

a. Interspecific hybrids were produced by crossing two *A. hypogaea* varieties (*hypogaea* and *vulgaris*) with three diploid species (*A. cardenasii*, *A. stenosperma*, and *A. batizocoi*). The hybrids had their fertility restored at the hexaploid level, and have been both self-pollinated and backcrossed to the respective cultivated parents. Pentaploids derived from two hexaploid generations have been self-pollinated to generate tetraploid progenies. DNA markers (RFLPs and RAPDs) have been screened to differentiate the cultivated and wild species. Progenies at all ploidy levels and hybrid generations will be evaluated for the presence of DNA from the wild species. This study will be used to determine the rate and timing of introgression from *Arachis* species into the cultivated genome and will enhance breeding efficiency of interspecific crosses which have previously expressed little apparent gene transfer between species.

b. Advanced generation hexaploid- and pentaploid-derived interspecific hybrids from crosses of A. hypogaea with 11 Arachis species will be propagated in the field to promote recombination among the cultivated and wild species genomes.

c. A molecular marker has been associated with nematode resistance. Attempts to find flanking markers will be made.

B. Philippines

1. Optimum sunlight and pH conditions

a. Seed production and promotion of UPLB-developed cultivars and promising materials (funded by P-CRSP project) at Laguna, Isabela, and Pampanga.

b. On-farm trials of promising peanut entries at Isabela, Pampanga, Zambales, Aurora Province, and other peanut-growing areas.

c. Monitoring performance of elite peanut lines entered in National Cooperative Tests across seven testing locations.

d. General yield trials at Laguna and Isabela.

e. Preliminary yield trials at Laguna and Isabela.

f. Advancement of segregating materials at artificial epiphytotic conditions and extraction of resistant materials at appropriate selection pressures.

g. Field screening for resistance to *Puccinia arachidicola*, *Sclerotium rolfsii*, viral diseases and *Aspergillus niger* wilt

2. Partial shaded growing condition

a. General yield tests of peanut entries intercropped under coconut at Quezon, Bicol, and other cooperating stations.

b. On-farm trials of shade-tolerant materials at various coconut testing sites such as PCA, Bago Oshiro, Davao City; Baao, Pili, Camarines Sur; Baler, Aurora Province, etc.

c. **Seed production of promising entries which are high yielding under partial shade and with extended shelf-life.**

3. **Highly acidic soil conditions**
   a. **Testing of advanced peanut lines at UPLB and Siniloan, Laguna**
   b. **Diallel cross for a genetic study on acid tolerance**

C. **Thailand: CRSP-funded activities were terminated December 31, 1994**

II. Training planned
A. US trainees
1. G. Garcia
   a. Degree: PhD
   b. Date of expected completion: July, 1995
   c. Country - present affiliation: US
   d. Thesis / dissertation topic: Introgression pathways of wild germplasm into cultivated peanut
2. G. Menon
   a. Degree: PhD
   b. Date of expected completion: June, 1996
   c. Country - present affiliation: US
   d. Thesis / dissertation topic: Development of transgenic peanuts with enhanced resistance to *Aspergillus* infection
3. S. Utomo
   a. Degree: PhD
   b. Date of expected completion: December, 1995
   c. Country - present affiliation: US
   d. Thesis / dissertation topic: Comparison of peanut plant regeneration techniques
4. C.A. Salas
   a. Degree: PhD
   b. Date of expected completion: June, 1996
   c. Country - present affiliation: US
   d. Thesis / dissertation topic: Evaluation of resistance to fungal foliar pathogens in Peruvian germplasm

B. Host Country trainees: None

III. Manuscripts, presentations, releases planned:
A. Manuscripts in process
1. Feng, Q. L., H. T. Stalker, and H. E. Pattee. n.d. Peanut plant regeneration through *in vitro* culture of peg tips of *Arachis hypogaea*. Crop Sci. (submitted 8/94).
2. Shew, B. B., M. K. Beute, and H. T. Stalker. 1994. Towards sustainable peanut production: Progress in breeding for foliar and soil-borne pathogens of peanut. Plant Dis. (accepted).
3. Stalker, H. T., G. A. Kochert, and J. S. Dhesi. n.d. Variation within the species *A. duranensis*, a possible progenitor of the cultivated peanut. Theor. Appl. Genet. (in review).
4. Tallury, Shyamalrau P., H. T. Stalker, and H. E. Pattee. n.d. Early reproductive ontogeny in interspecific crosses of *A. hypogaea* and section Arachis species. Ann. Bot. (in review).
5. Utomo, S. D., A. K. Weissinger, H. T. Stalker, and T. G. Isleib. n.d. High efficiency of peanut regeneration using a nonimbibed immature leaflet culture method. Peanut Sci. (in review).
6. Waliyar, F., B.B. Shew, R. Sidahmed, and M.K. Beute. n.d. Effects of host resistance on germination of *Cercospora arachidicola* on peanut leaf surfaces. Peanut Sci. (in preparation).

B. Presentations
1. Anderson, Stalker, Grignon, Shew, and Beute. Resistance to white mold (*Sclerotium rolfsii*) within wild peanut accessions. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.

2. Barrientos-Priego, Wagner, Elkan, Isleib, and Pattee. Evaluation of *Bradyrhizobium* isolates from soil samples obtained from pods of Mexican *hirsuta* type landraces. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
3. Brune and Beute. Root growth responses of peanut genotypes following mechanical wounding to simulate damage by the pathogen *Cylindrocladium parasiticum*. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
4. Butzler, Bailey, and Beute. Efficacy of fluazinam applications and canopy alterations (mechanical and phenotype) on Sclerotinia blight incidence. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
5. Feng, Stalker, and Pattee. Plant recovery in *Arachis* by *in vitro* culture of peg tips, ovules, and embryos. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
6. Ferguson and Beute. Effect of straw amendment on incidence of disease caused by three soilborne pathogens of peanut. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
7. Randall-Schadel, Bailey, Beute, and Dowell. Potential use of optical scanners to separate seed with a testa symptom associated with seedborne *Cylindrocladium parasiticum*. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
8. Shew, Beute, and Isleib. Resistance to Sclerotinia blight and Southern stem rot in breeding lines of virginia peanut. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
9. Shew, Beute, Stalker, and Isleib. Durable disease resistance in peanut. 13th Intl. Plant Protection Congress, July 2-7, 1995, The Hague, Netherlands.
10. Stalker, Shew, Garcia, Beute, Barker, Holbrook, Noe, and Kochert. *Meloidogyne arenaria* resistance in advanced-generation *Arachis hypogaea* x *A. cardenasii* hybrids. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
11. Utomo, Weissinger, Stalker, and Isleib. Comparison of somaclonal variation caused by three peanut regeneration methods. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.
12. Weissinger. Transgenic plant protection strategies. 1995 Amer. Peanut Res. Educ. Soc., July 11-14, Charlotte, NC.

C. Releases
1. Release of CBR-resistant NCSU breeding line N90013E (pending approval by releasing institutions)

IV. International travel planned
A. R. Abilay
1. Month(s): July, 1995
2. Destination: APRES annual meeting, Charlotte, NC
3. Duration: 1 week
B. T.G. Isleib
1. Month(s): April, 1996
2. Destination: UPLB, Los Baños, the Philippines
3. Duration: 1 week

V. Project redirections proposed: None

TX/MM/S,G

Mycotoxin Management in Peanut by Prevention of Contamination and Monitoring

Texas A&M University, College Station, Texas
Institut Senegalais de Recherches Agricoles, Bambey, Senegal
Institut de Technologie Alimentaire, Dakar, Senegal
University of Science and Technology, Kumasi, Ghana
Food Research Institute, Accra, Ghana

Princicpal Investigators:
TAMU: Marian N. Beremand, Nancy P. Keller, Timothy P. Phillips
ISRA: Amadou Ba
ITA: Amadou Kane
UST: Richard T. Awuah
FRI: Kafui Kpodo

INTRODUCTION

Mycotoxin contamination of peanut poses a serious health hazard to consumers of peanut products throughout the world. Effective mycotoxin management in peanut depends upon the implementation of multiple strategies to prevent contamination or reduce levels of aflatoxins, and other mycotoxins, which may be present in peanut and peanut derived products. This project is designed to conduct experiments which can further identify the mycotoxins and mycotoxigenic fungi present on peanut; define compounds from peanut and/or other organisms that prevent mycotoxin accumulation; implement molecular biological tools to understand the biosynthesis of mycotoxins at the gene level; select peanut germplasm with resistance to aflatoxin production; improve agronomic production practices which reduce the levels of contamination within the field; improve storage practices which minimize further contamination; develop rapid, practical and economical monitoring procedures for the detection of mycotoxins; and devise cleaning, sorting, and detoxifying protocols for removal or deactivation of mycotoxins within peanut and/or peanut derived products.

IMPACTS

Host Countries - Reducing the levels of aflatoxin/mycotoxins in peanuts will help improve the health of the local population, and improve the quality of peanut products. New methods of aflatoxin detection should aid in quantifying levels of aflatoxin present, and aid in justifying the diversion of contaminated peanut into processing for clean-up and/or detoxification. Development of improved marketing procedures, those which provide an incentive for producing aflatoxin-free peanut, will help encourage implementation of aflatoxin preventive measures and removal of mold-damaged seed previous to processing. Newly developed aflatoxin sorption methodologies will allow local villagers and industry to treat peanut oil to reduce aflatoxin levels to a safe level for consumption. Continued screening of peanut genotypes for resistance to penetration by the mycotoxin producing fungi will further help reduce the degree of contamination of peanut at the village level. Identification of local plants which exhibit fungistatic properties or inhibit mycotoxin production can be used in biocontrol programs. Training efforts will improve technology transfer to host countries and be instrumental in the much needed development of molecular biology programs at universities and research institutes. Finally, successful research in the above areas will be applicable to other crops affected by mycotoxins.

In the United States - Research results will lessen the impact of the mycotoxin problem to the peanut industry in the U.S. Molecular and genetic analyses of Aspergillus and Fusarium mycotoxin genes offer new approaches to preventing mycotoxin contamination and for selecting or producing mycotoxin resistant peanut cultivars. Identification of 'anti-Aspergillus/anti-aflatoxin' genes could lead to transgenic peanut cultivars. Newly developed aflatoxin resistant peanut cultivars, adapted to the peanut growing regions of the United States, could greatly reduce the number of Segregation III peanut marketed within the country. Marketing of peanut seed with reduced levels of mold and mycotoxin damage will reduce the costs of processing peanut products. Improved mycotoxin detection techniques should increase the speed and accuracy of analyses, and reduce the costs related to the

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diversion of contaminated peanut lots. Newly discovered sorption techniques to bind and inactivate aflatoxins, and other mycotoxins should increase the margin of safety to insure that these products are safe for use as food or feed components.

MAJOR ACCOMPLISHMENTS

Both Dr. Richard T. Awuah (plant pathologist from UST, Kumasi, Ghana) and Ms. Kafui Kpodo (food biochemist from FRI, Accra, Ghana) visited Texas A&M this past year. Dr. Awuah won a biotechnology training grant from UNESCO and spend 6 weeks in Dr. Keller's laboratory learning molecular biological techniques. Ms. Kpodo received training from Dr. Timothy Phillip's lab on identifying sterigmatocystin, fumonsin and other fungal mycotoxins. Ms. Kpodo also visited with Dr. Elena Castell-Perez at Alabama A&M, Huntsville, AL. The complimentary interactions between the Mycotoxin and Food Utility CRSPs should result in more efficient use of resources.

The first groundnut survey in Ghana showed that aflatoxin was detected in all the damaged kernel samples. Most of these samples were characterized by aflatoxin levels in excess of the WHO recommended value of 30 ppb. Aflatoxins were not detected in some of the undamaged kernel samples and even when present in such kernels, the levels were generally very low.

Microbial assays using colored aflatoxin mutants in Aspergillus flavus and Aspergillus parasiticus have continued. Collaborative work with Dr. Olin Smith and graduate student Yolanda Lopez have demonstrated that the use of the norsolorinic acid mutant (red color) may give superior results than a wildtype aflatoxin producing strain of Aspergillus in analyzing aflatoxin contamination of peanuts.

Co-operative studies with Dr. Thomas H. Adams (Department of Biology, Texas A&M University) have resulted in identification of the entire sterigmatocystin (ST) pathway of Aspergillus nidulans. Evidence shows that the sterigmatocystin pathway is functionally equivalent to the aflatoxin (AF) pathway in A. flavus and A. parasiticus and that this ST/AF toxin pathway is conserved as a cluster of genes in all producing aspergilli. Most importantly, the regulatory gene, aflR, is found to regulate both the sterigmatocystin and the aflatoxin pathway in all three Aspergillus spp. Efforts will be focused on identifying ways to keep aflR turned off permanently. The study of this and other genes in the ST/AF pathway will lead to a molecular understanding of what environments, peanut metabolites, etc. result in aflatoxin production in peanuts.

Identification and characterization of the genes in Fusarium species for trichothecene biosynthesis has revealed the presence of a new regulatory region within the trichothecene gene cluster. This regulatory region, which encompasses a DNA sequence that appears to encode for a protein, designated Tri10, is involved in the coordinated expression the genes required for trichothecene biosynthesis. DNA sequence date has shown that the predicted Tri 10 protein sequence contains a nuclear transit signal characteristic of proteins located in the nucleus. Experiments are underway to determine if disruption of Tri 10 will prevent or increase trichothecene synthesis. This DNA region may prove to be a key control point for preventing trichothecene toxin production.

GOALS

The goals of the mycotoxin management project are to develop and apply techniques which reduce the severity of the mycotoxin contamination problems within the Lesser Developed Countries and the United States. Prevention of mycotoxin contamination through the development of aflatoxin resistant cultivars, effective biocontrol strategies, and improved production, harvest, storage, and processing procedures will help reduce the costs of excessive processing to remove the mycotoxins. We will utilize the most appropriate technologies from biotechnology to cultural practices as best fit. For those peanuts which still contain mycotoxins the goal is to develop improved inspection and diversion procedures, and clean-up and detoxification procedures, which will reduce the levels of mycotoxins present within peanut and peanut products to assist in meeting governmental health and safety standards.

OBJECTIVES

1. Investigate the genetic and molecular mechanisms that control mycotoxin biosynthesis in Aspergillus and Fusarium spp.
2. Develop new assays and studies to assess the importance of other mycotoxins and mycotoxigenic fungi in peanut.
3. Assess Ghanaian plants and plant extracts for their ability to decrease Aspergillus growth and/or aflatoxin production.
4. Identify single gene products which inhibit Aspergillus growth and/or aflatoxin production and use them to transform peanut plants.

ORGANIZATION AND PERSONNEL

U.S. Lead Institution:
Texas A&M University (TAMU)

Principal Investigator
Dr. Marian N. Beremand, Department of Plant Pathology and Microbiology (PPM), TAES, TAMU, CS

Co-Principal Investigators
Dr. Nancy P. Keller, Department of Plant Pathology and Microbiology, TAES, TAMU, CS
Dr. Timothy D. Phillips, Department of Veterinary Public Health (VPH), TAMU, CS

Cooperators
Dr. Olin D. Smith, Department of Soil and Crop Sciences, TAMU, CS
Dr. Thomas H. Adams, Department of Biology, TAMU, CS
Dr. Charles Simpson, Department of Soil and Crop Sciences, TAMU, CS

Technician
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Ms. Gennie Schuller, Department of PPM, TAES, TAMU, CS

Graduate Student
Mr. Andrew Tag, Department of PPM, TAES, TAMU, CS

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Dr. ulnara Garifullina, Department of PPM, TAES, TAMU, CS

Budget Analyst
Ms. Judy Owens, TAMU Research Foundation

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Dr. Neal Van Alfen, Head, Dept. of PPM, TAES, TAMU, CS

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Directeur General
Dr. Mouhamadou Diop, ITA, Dakar-Hann, Senegal, B.P. 2765

Ghana: University of Science and Technology (UST)
Principal Investigator
Dr. Richard T. Awuah, Department of Crop Science, UST, Kumasi, Ghana

Ghana: Food Research Institute (FRI)
Ms. Kafui Kpodo, FRI, Accra, Ghana

APPROACH

Determine the molecular mechanisms that control mycotoxin production by utilizing the techniques of molecular biology and genetics to identify and clone the structural and regulatory genes required for toxin biosynthesis in Aspergillus and Fusarium species.

Using microbial assays to identify cultural conditions which may inhibit or induce ST/AF production in Aspergillus spp.

Identify Ghanaian plants which inhibit Aspergillus growth and/or ST/AF production.

Evaluate and further develop mycotoxin detection systems and detoxification methodologies for mycotoxin contaminated peanut and peanut products.

Further assist, train and encourage host country scientists in conducting, developing and establishing their own mycotoxin research programs and in attaining project goals, including the transfer and adaptation of technologies developed in conjunction with this research project.

RESEARCH ACCOMPLISHMENTS

Texas

Use of microbial assays in evaluation of peanut cultivars and genotypes for Aspergillus colonization and aflatoxin production

Studies were continued on evaluating a new microbiological assay to assess factors which regulate aflatoxin production and to test peanut cultivars for relative resistance/susceptibility to Aspergillus infestation and/or aflatoxin contamination. This assay utilizes Aspergillus aflatoxin mutants which accumulate colored intermediates in the aflatoxin pathway The norsorolinic acid - red color - mutant has been most extensively utilized. This work has benefited by cooperative studies with Dr. Olin Smith and Ms. Yolanda Lopez who have used the norsolorinic mutant for Ms. Lopez' Master's thesis. Ms. Lopez' results indicate that this assay is useful in identifying peanut genotypes which reduce fungal growth and AF biosynthesis.

Inducing and Inhibitory Environments for Aflatoxin Production

Use of the norsolorinic acid mutant have also been helpful in identifying potential enviroments which may inhibit or induce AF production. Preliminary studies indicate that the pH of the environment effects AF production.

Molecular biology and genetics of mycotoxin production

In a collaborative project, Dr. Keller and Dr. Adams identified what appears to be the entire ST pathway in A. nidulans. Most importantly, the ST pathway is functionally and structurally conserved with the AF pathway in A. flavus and A. parasiticus, e.g. these pathways are nearly the same which implies that the same control method working to reduce AF will also reduce ST production. The progress in this molecular project significantly contributes to the Peanut CRSP project; eventually any inhibitors/inducers of ST/AF will be assessed for their effect on specific genes in the aflatoxin pathway. We continued with characterization of the Aspergillus nidulans verA gene by studying the sequences in the promoter necessary for gene expression. The gene encodes for a reductase activity thought to be active in the aflatoxin pathway between versicolorin A and sterigmatocystin.

Studies have been initiated to obtain mutants of Fusarium moniliforme that are altered in fumonisin production. During the past year, immunospecific screening of UV-treated colonies for fumonisins provided a collection of potential mutants. We will continue to characterize these mutants and identify those which are specifically blocked in either the fumonisin biosynthetic or regulatory pathways. Such mutants will be used to map and clone the structural and regulatory genes that control the production of these mycotoxins and to assess the role of fumonisins in the infection of peanuts (and other crops) by F. moniliforme.

Significant progress has been made towards identifying key regulatory controls governing the synthesis of the trichothecene mycotoxins by F. sporotrichioides. Characterization of a novel group of transformants has provided genetic evidence for a region within the trichothecene gene cluster that is responsible for coordinately shutting off transcription of the other known structural and regulatory genes located in the trichothecene gene cluster. Experiments are being conducted to determine the mechanism by which this DNA region is controlling the expression of the trichothecene genes. This work is being conducted in a new collaborative project between Dr. Beremand and Dr. Hohn.

Phyllosilicates and Clays

In a collaborative study, Dr. Phillips and scientists at Prairie View A&M University and the USDA/ARS in College Station, have recently shown that hydrated sodium calcium aluminosilicate (HSCAS) can significantly reduce the levels of aflatoxin M1 in milk from lactating dairy goats exposed to 100 and 200 ppb aflatoxins in the diet. A marked reduction in the aflatoxin content of milk (52-87%) resulted from the dietary addition of 1 to 4% of the HSCAS (w/w). No differences in feed intake, milk production or milk quality were observed in the presence of high levels of HSCAS (i.e., 4%) or HSCAS + aflatoxins.

Mycotoxin detection assays

Work in Dr. Phillips laboratory has resulted in the construction and evaluation of a minicolumn method for the field-practical detection of fumonisins in corn. Collaborative work with Dr. Beremand is in progress to optimize this simple test for the detection of fumonisins in peanut. Also, a modified HPLC procedure has been recently developed for the analytical separation, detection and quantitation of fumonisins in peanut. This newly developed method will be utilized to confirm the accuracy and sensitivity of the fumonisin minicolumn.

Dr. Phillips is also evaluating another minicolumn assay developed at Texas A&M University for the rapid screening ofmilk and dairy products for aflatoxin M1. The limit of detection for this assay has been found to be very low (i.e. 0.2 ppb) and is comparable to commercially available immunoaffinity assays. This new test appears to be accurate, rapid, easy to perform and chemically stable.

Senegal ISRA

No activity was reported.

Senegal ITA

No activity was reported.

Ghana UST and FRI

Groundnut samples stored for 6-8 months were collected from markets in each of the ten regions in Ghana. Samples from a market were bulked together, sorted out into undamaged and damaged kernels, and the percentage of damaged kernels estimated. Fungi associated with the kernels were assayed on half strength potato-dextrose agar and identified using appropriate texts. Aflatoxins were extracted from the kernels following the method of Pons. They were identified and quantified by reversed phase liquid chromatography with post column iodine derivatization.

Results:

The extent of damage in groundnut sample collected from all ten regions in Ghana ranged from 1.5 to 9.25% (Average 3.8%). Of the two aflatoxigenic fungi, A. flavus was more frequent, being observed from damaged kernels with a frequency of 31.7% and from undamaged kernels 12.8% of the time. Aspergillus parasiticus was not as commonly encountered and was absent from 37% of the 42% groundnut samples assayed.

A. niger van Thieghem was recorded in high frequencies on both healthy (29%) and damaged (39%) kernels. Metabolites from this fungus have occasionally been shown to be toxic, so the isolates of A. niger from groundnut may be useful to analyze for mycotoxins in the future.

Other fungi encountered on both damaged and unhealthy kernels, although in extremely low frequencies, were A. candidus Link, A. tamarii Kita, A. ochraceous Wilhelm, Penicillium spp, a Trichoderma sp, Rhizopus stolonifer, and a Mucor species. A few unidentifiable fungal species also were encountered. Some of these fungi have been reported as residents of stored peanuts (Joffe and Borut, 1966) and are capable of producing toxins other than aflatoxins.

Aflatoxin was detected in all the damaged kernel samples. Most of these samples were characterized by aflatoxin levels in excess of the WHO recommended value of 30 ppb. Aflatoxins were not detected in some of the undamaged kernel samples and even when present in such kernels, the levels were generally very low.

Conclusions:

The groundnut aflatoxin problem in Ghana may be partly solved by sorting out damaged kernels from undamaged ones - which should not be difficult to do since the extent of damage to groundnut in storage is low.

Assessment of Plant Extracts for anti-Aspergillus and anti-aflatoxin properties.

Eighteen plant extracts were tested for possible anti- Aspergillus and anti- aflatoxigenic effects. Extracts were prepared as steam distillates, hot water infusions, and pure oils. The assay was accomplished on two aflatoxin inducing media, viz Yeast Extract Sucrose (YES) medium and 1/2 strength Potato Dextrose Broth (PDB). Four milliliters of media contained in screw capped vials were amended with 0.2 ml of each plant extract, inoculated with a norsolorinic Acid mutant of A. parasiticus (Keller et al. 1994) and incubated for 8 days.

Results:

Steam distillates from Xylopia aethiopica (Dunal) Rich, Monodera myrstica (Gaerth), Cinnamomum verum Presl. and hot water infusion from Piper nigrum L. completely inhibited NOR formation in PDB, but these extracts were ineffective in the YES medium. In preliminary experiments, however, we have noted that at concentrations of 0.5 ml and above /4 ml YES medium, distillates from Ocimum gratissimum L., Cymbopogon citratus (D.C.) Stapf. and Xylopia aethiopica completely prevented growth of A. parasiticus. This agrees with a previous report of fungitoxity of extracts from these three Ghanaian traditional medicinal plants (Awuah, 1989). Extracts from these plants as well as those from C. verum and Syzygium aromaticum, which also completely inhibited fungal growth in the present study, should be considered for use in treating groundnut kernels to control the aflatoxin producing fungi.

PUBLICATIONS AND PRESENTATIONS

Publications

1. Lopez Y, Smith O D, Keller N P (in preparation) The use of colored Aspergillus mutants to screen for aflatoxin production in peanuts. Crop Science
2. Yu J, Butchko R A E, Fernandes M, Keller N P, Leonard T, Adams T (submitted) Conservation of structure and function of the aflatoxin regulatory gene aflR from Aspergillus nidulans and A. flavus.. Curr Gene
3. Keller N P, Segner S, Bhatnagar D, Adams T H (in press) stcS encodes a putative P450 monooxygenase needed for the conversion of versicolorin A to sterigmatocystin in Aspergillus nidulans. Appl Environ Micro.
4. Keller N P, Adams T H (1995) Analysis of a mycotoxin gene cluster in Aspergillus nidulans. In S.A.A.S. Bulletin-Biochem. Biotech. (in press)
5. Keller N P, Brown D, Butchko R A E, Fernandes M, Kelkar H, Nesbitt C, Segner S, Bhatnagar D, Cleveland T E, Adams T H (1995) "A conserved polyketide mycotoxin gene cluster in Aspergillus nidulans." In Proc. from UJNR: Molecular Approaches to Food Safety Issues Involving Toxic Microorganisms, J L Richard Ed., Chapter 18.

Presentations and Workshops

Drs. R. Awuah, A. Ba, M. Beremand, A. Kane, N. Keller, T. Phillips and Mrs. K. Kpodo all attended the Peanut CRSP meeting in Arlington the end of March.

Dr. N. Keller attended the USDA Aflatoxin Elimination Workshop end of October 1994 in St. Louis, Mo.

Drs. M. Beremand and N. Keller attended the UNJR Joint Toxicology Meeting in Peoria, Ill in Nov. 1994.

Drs. M. Beremand and N. Keller attended the 18th Fungal Genetics Conference in Asilomar, CA in March 1995. Dr. Beremand served as the organizor for the workshop on Secondary Metabolites: Gene organization and Regulatory Mechanisms. Both Drs. Keller and Beremand gave invited presentation at the workshop and presented posters at the meeting.

Drs. M. Beremand and N. Keller attended the Gordon Mycotoxin/Phycotoxin Conference in June. Dr. Keller presented a talk entitled "Conservation of a Mycotoxin Gene Cluster in Aspergillus nidulans"

TRAINING: DEGREE AND SHORT-TERM

Dr. Richard T. Awuah completed a training session in Texas A&M University Sept - Oct 94 as previously described.

Ms. Kafui Kpodo complete a training session at Texas A&M University in April 1995.

PLANS FOR 1995-1996

1. A complete revision of the Peanut CRSP requires new proposals.
2. Dr. Awuah and Ms. Kpodo will be attending a African-wide Mycotoxin Conference in Cape Town, RSA in January 1996.

NCS/IM/TP

Management of Arthropods on Peanut in Southeast Asia

NORTH CAROLINA STATE UNIVERSITY
KHON KAEN UNIVERSITY
DEPARTMENT OF AGRICULTURE, THAILAND
UNIVERSITY OF THE PHILIPPINES, LOS BANOS

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INTRODUCTION

The uncertainty of current peanut price support legislation and consumer concerns over pesticide use in the U.S. and increasing demand for a high protein food source produced economically in developing countries demands continued research focused at low-input, sustainable systems. This research integrates past objectives toward reduced pesticide use practices in peanut production and are the result of world-wide efforts to meet the objectives of integrated pest management. The acquisition of the data base required to reach these goals is the result of well-balanced and strong basic and applied research components for insect management. A thorough understanding of the biological, economic, and social factors involved is also necessary. The production of peanuts throughout Southeast Asia is a complex agroecosystem subject to a wide variety of plant stresses as well as certain socioeconomic factors. A rainy and a dry season crop are grown in many areas each year and insect pests cam be a limiting factor for peanut yield and quality. The management of several insect pests pose a significant challenge for peanut producers. The whole plant may be attacked by one or more species and the plant is exposed to potential insect feeding virtually the whole growing season However, soil insects provide the greatest management challenges. Not only do these pests feed directly on the harvested unit, the pod, and result in considerable crop loss, but the subterranean nature of the pests renders them difficult to detect and manage. To effectively manage these pests in an economically and environmentally-sound approach lending itself toward sustainable agriculture, a sound biological base must be established. Research needs for all insect pests include ecological studies, surveys of pest problems, damage assessments and thresholds, population monitoring and scouting techniques, off-target impacts of pesticide use and potential interactions. A critical component of the sustainable production package is the development and implementation of alternative control strategies including host-plant resistance, cultural and biological control. Such broad-based research programs are difficult due to the field nature of the research, the breadth of the problems to address, and the resources required to effectively conduct such efforts. The information that results, however, helps build effective integrated pest management programs and moves peanut production closer to the goal of a sustainable production system for peanut production in all countries.

IMPACTS

Despite significant progress in cultural practices, alternative control strategies, and host-plant resistance, pesticides continue to play a major role for managing insect pests in peanuts in many countries. In the United States and in Southeast Asia, pesticide use is considered an essential tool for preventing significant crop loss from foliar feeding pests. These situations require an improved system to aid the grower decision making process for insecticide use to limit the indiscriminant use of pesticides while seeking more sustainable, alternative means of pest control. Such information on thresholds and biorational control approaches is being refined in the Philippines and Thailand as well as the U.S. Studies have documented the benefits of well-timed treatments based on thresholds and the use of biological materials such as *Bacillus thuriengensis*, neem seed extract, and NPVs. Growers in Southeast Asian countries who have observed these trials are accepting the use of thresholds (where available) and prescription pesticide use rather than calendar-type sprays.

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The past efforts in the Philippines and Thailand spent evaluating varietal tolerance and resistance of new genotype entries are just now beginning to pay dividends. Cultivars with more tolerance or resistance(including currently recommended varieties) are being evaluated in IPM programs grown in the actual production areas to test their overall role as integrated into a complete management system. The significant effect of natural enemies and inundative releases of parasites has been documented in several areas of the Philippines and proven quite successful and economically sound. This is only possible however, in a reduced pesticide use system. Efforts in Thailand has produced a clearer understanding of the relationship between various sucking insects and the presence of certain virus diseases that they vector. Researchers in both the Philippines and Thailand have continued to develop crop loss assessment data for several pests despite the inherent difficulty associated with the development of such information. Finally, studies in both countries have documented more closely the impact of soil insects on peanut production and have produced valuable insight into the biology, ecology, and possible control options for these pests.

In North Carolina efforts to minimize pesticide use has been frustrated by the arrival of new pest problems, such as tomato spotted wilt virus which is vectored by thrips. However, peanut growers have dramatically improved their ability to use prescription applications of pesticides for certain pests. This includes the use of more environmentally-sound pesticide selection and more efficient timing of application. These improve control strategies generally reduce overall pesticide use and reduce costs while preserving crop yield and quality. Two empirical models for predicting pest outbreaks continue to prove useful in alerting growers to potential pest outbreaks. Work with cultural practices have demonstrated tillage can reduce pest population levels and minimum tillage concepts are gaining some acceptance from growers.

North Carolina growers continue to use the economic thresholds for thrips, corn earworms, and leafhoppers in peanut IPM programs. These allow growers to avoid some pesticide applications and to make more intelligent decisions in light of the potential threat of tomato spotted wilt virus vectored by thrips. The use of alternate, less toxic products for thrips management has been demonstrated to have economic as well as environmental advantages. Extensive field testing of 84 different entries in a host-plant resistance has provided promise for new opportunities in insect management using resistant germplasm. New information concerning thrips and virus overwintering is useful in developing a management approach for this problem. Significant varietal affects on virus incidence in the host-plant resistance plots indicate some potential for the use of varieties to aid in an overall management strategy for this serious problem.

MAJOR ACCOMPLISHMENTS

PHILIPPINES

Researchers in the Philippines have made outstanding progress in evaluating host-plant resistance, biological control, alternative management strategies, developing economic thresholds, and pest biology. The progress seen in the host plant resistance work has developed an excellent data base evaluation under Filipino production systems. The principal investigators have taken the next important step and are testing the integration of this information into IPM programs in the various production regions. Studies investigating these IPM systems are underway in the Cagayan Valley, near Pamapanga, and in Mandaue City. The data from these trials have shown that overall insect damage in IPM plots is less than in standard "farmer managed" plots and conventional insecticide use was often avoided. Some additional studies have looked at current Seed Board recommended varieties and promising cultivars used in conjunction with classical biological control programs.

Insecticides use is still necessary in some areas and these have proven effective against many pests. Biological materials such as *Bacillus thuriengensis* have proven very effective against a complex of defoliators. These studies and those on the effects of intercropping and fertilizer use have contributed greatly to the development of effective IPM programs.

The continuation of studies validating the use of biological control programs using *Trichogramma* sp. and a microbial insecticide *Bacillus thuringiensis* shown promise for future wide-scale implementation. Biological control studies have shown practicality and profitability using an integrated program in IPM evaluation trials.

THAILAND

The researchers in Thailand continue to place primary emphasis in this program at the evaluation of entries for insect resistance at both DOA and KKU. Studies continued to investigate the development of a variety of control strategies for IPM programs. This included basic insecticide evaluations to improve the timing of application to increase yield and total integrated program using host plant resistance as well as cultural and biological control methodologies. Since insecticides are relied upon quite heavily in certain areas these studies are essential. Studies on the relationship of virus incidence and transmission has been limited in the past year as new techniques to determine this relationship are being defined. These past studies have enhanced our understanding of the dispersal of several insect pests and the risk of disease transmission.

New studies in Thailand have investigated the relationship of pod-feeding insect pests and the incidence of aflatoxin. Since recent surveys have documented the widespread nature and seriousness of soil insect pests of peanuts in Southeast Asia, this work is critical in developing a complete understanding of crop loss assessment. It is also important in future aflatoxin elimination management strategies.

NORTH CAROLINA

Emphasis on the development of a sustainable agricultural system for peanuts continues to be a major objective for the U.S. project. Increased effort to minimize preventive pesticide use and to integrate biological and cultural control practices have been expanded. The use of biological control products is showing increasing promise for insect management in peanuts. These studies closely correlate similar studies in the Philippines. The validation of forecast models for the lesser cornstalk borer and spider mites has proven to be an invaluable tool for pest management. Evaluations of pesticide interactions are increasing our understanding and provide us with an approach to reduce overall pesticide dependence.

Studies have been concluded and provide us with an approach on the wide-area thrips dispersal and migration, thrips' overwintering, and within the field dispersal. New studies are focusing more on the overwintering of the thrips and the tomato spotted wilt virus. These were conducted to determine virus/vector relationships with the increasing threat from tomato spotted wilt virus in peanuts which is vectored by these thrips. In addition, new studies looking at varietal difference in the incidence of this virus are underway. These seek to determine if these differences are the result of inherent tolerance or resistance to the virus by the plant or some effect on thrips feeding behavior by different cultivars. Upon completion, these studies will dramatically improve our understanding of the vector and help to manage this disease. These studies complement past vector/virus relationship work in Thailand.

GOALS

The major goal of this project is the effective management of those arthropod pests that limit peanut production through an effective pest management program based on sound principles of IPM and sustainable agriculture.

OBJECTIVES

1. To continue the evaluation of a wide variety of genetic material for possible insect tolerance or resistance to single species and arthropod complexes.

2. To develop damage assessment data for the arthropod complex and relate to host plant phenology. This information will be incorporated into economic thresholds for use in IPM programs.

3. Study the biology and ecology of the important arthropod pests. This includes studies on virus/vector relationships. Conduct surveys to determine abundance and pest status of arthropods.

4. Determine the effect of cultural practices (i.e., tillage) on insect populations and host plant damage.

5. Utilize monitoring devices to gain a better understanding of insect biology and use as a predictive tool for insect occurrence and pest abundance.

6. Develop an effective IPM program and demonstrate the benefits through research findings and on-site demonstrations utilizing information developed from the Peanut CRSP in North Carolina, Thailand, and the Philippines.

7. Evaluate the potential for biological control as a realistic approach to arthropod management in peanuts.

ORGANIZATION

A.	U. S. Lead Institution:	North Carolina State University
	Principal Investigator:	Dr. Rick L. Brandenburg, Dept. of Entomology
	Technicians:	Mr. Peter T. Hertl, Dept. of Entomology
		Mr. Brian Royals, Dept. of Entomology
		Ms. Brenda Watson, Dept. of Entomology
	Graduate Students:	Mr. Rob Hummel, Dept. of Entomology
		Mr. Lloyd Garcia, Dept. of Entomology
	Cooperators:	Dr. Tom Isleib, Dept. of Crop Science
		Dr. Tom Stalker, Dept. of Crop Science
		Dr. Jack Bailey, Dept. of Plant Pathology
B.	Counterpart Institution:	Khon Kaen University, Khon Kaen, Thailand
	Principal Investigator:	Dr. Manochai Keerati-Kasikorn, Dept. of Entomology
	Cooperators:	Sunisa Sakulleerungrode, Dept. of Entomology
		Methee Rohitasathien, Dept. of Entomology
	Technician:	Mr. Preecha Singha, Dept. of Entomology
C.	Counterpart Institution:	Department of Agriculture, Field Crops, Bangkok, Thailand
	Principal Investigator:	Turnjit Satayavirut, Entomology & Zoology Division
	Cooperators:	Ms. Srisamorn Pitak, Entomology & Zoology Division
	Technician:	
	Mr. Punya Pooksoon,	Entomology & Zoology Division
D.	Counterpart Institution:	University of the Philippines at Los Banos, Philippines
	Principal Investigator:	Dr. Virginia Ocampo, Dept. of Entomology
		Dr. Eliseo Cadapan, Dept. of Entomology
	Cooperators:	Dr. Edwin Benigno, National Crop Protection Center
		Mr. R. V. Ebora, National Inst. of Biotechnology
		Mr. Dante Santiago, Dept. of Entomology
		Dr. Remedios M. Abilay, Institute of Plant Breeding
	Research Associates:	
		Mr. Octavius Caesar P. Ebron, Dept. of Entomology
	Graduate Students:	Teddy de la Rosa, Dept. of Entomology
		Lorene Trapego, Dept. of Entomology
E.	USAID Project Officers:	K. A. Rushing/Manila, Philippines

APPROACH

Principal investigators in the Philippines, Thailand, and North Carolina will continue to investigate various peanut genotypes. These genotypes are being grown in several locations within the peanut-producing regions to provide data under a range of agronomic, environmental, and insect pest conditions. Since multiple species of insects attack peanuts in Southeast Asia, the search for resistance to insect guilds rather than a single species is emphasized. Thus broad resistance would probably result in a more sound release for growers, since the off-target effects of host plant resistance on related pest species is not well understood. Through many years of testing with varying levels of pest pressure and different environmental factors several entries have shown promise. These will continue to be studied and plot size expanded to ensure true tolerance or resistance exists and damage is not simply a reflection of preference. The more suitable entries are being tested in advanced IPM trials to determine the cultivars that show the best fit into such systems for the most integrated overall approach. In North Carolina, the host-plant resistance effort was recently expanded in cooperation with Dr. Tom Isleib with renewed effort on foliar feeding insect pests.

Surveys of all arthropod pests, especially subterranean ants and other soil pests, will continue in Southeast Asia. These studies are being enhanced with the addition of studies on the biology, ecology, and the management of the subterranean pests. Additional research on sampling and traditional management approaches is critical for soil pests and will continue, but such research is slow to produce results. Very little is know about crop losses to these and other pests. Insect abundance and plant damage relationships are being refined through the continuation of crop loss assessment studies. New efforts on developing a better understanding of the relationship of thrips movement and overwintering and the incidence of the tomato spotted wilt virus continue at an increasing pace. Related studies on vector and virus relationships will be evaluated in Thailand. Traps to monitor migration, as well as studies of feral host plants, insect abundance in field, and disease incidence, will be monitored. The continued increase in the incidence of tomato spotted wilt virus in the North Carolina peanut crop each year has justified a more significant effort investigating the role *Frankliniella fusca* laying in vectoring this disease in peanut. In North Carolina, these studies will focus exclusively on thrips, while in Thailand, studies will include aphids and jassids as well. The effect of varieties on the incidence of the disease will be closely examined in North Carolina.

The demand for low input systems worldwide and reduced pesticide use in the U.S. encourages a continuation of studies on the use of biological control agents in the Philippines and North Carolina. These studies will include the release of egg parasites, *Bacillus thuringiensis*, nuclearpolyhedrosis virus,and entomogenous nematodes, with the ultimate goal of incorporating these into a totally integrated control system. Since pesticides continue to be an important tool in the management of insect pests of peanuts, studies looking at the off-target effects of pesticides will be continued in all three countries to determine the detrimental effects and the potential to induce secondary pest problems.

The past five years of research have developed the essential components of an Integrated Pest Management program in each respective country. Each of these programs vary significantly in their comprehensiveness and management tools used, but each forms a bridge toward a more sustainable approach. Further research is targeted toward more refined programs. Due to the changing emphasis within USAID and the Peanut CRSP, the overall objective in Thailand is to bring together the findings from the many years of research and system validation and begin wide-scale implementation through the Department of Agriculture Extension. Furthermore, the research expertise and accomplishments will be directed to function as a regional center of excellence for peanut pest management in training scientists from surrounding countries throughout Southeast Asia.

ACCOMPLISHMENTS IN DETAIL

PHILIPPINES - Drs. Ocampo and Abilay continue to coordinate the host-plant resistance research effort and this has resulted in an excellent joint program. The data base developed by Dr. Ocampo has provided an excellent overview of the resistance available for the pest species in the Philippines. The recent expansion of this program to the research station in the Cagayan Valley region has been a much need effort to further validate the usefulness if these genotypes. The importance of peanuts in this region and the different pest pressures there make

this work essential to the eventual widespread adoption of new technologies and varieties. This work is made possible through the superb cooperation of the superintendent at this station, Val Perdido. Dr. Cadapan focused exclusively on genotype evaluations during 1994-1995. This work did include the use of various IPM strategies across certain cultivars and work was conducted in the Zambales region. Dr. Ocampo has continued her focus on appropriate insecticides and intercropping peanut with banana and other crops as well as the influence of fertilizer on pests outbreaks.

The studies conducted by Dr. Ocampo both at UPLB and in the north at Cabagan, Isabela and San Marcos, Quirino focus on host plant resistance. New studies are underway at the Cagayan Valley Agricultural Experiment Station and offer a good opportunity for the evaluation insect resistant cultivars and testing their integration into local production practices. The research done at this location serves as an excellent educational tool to assist technology transfer once practical information is developed.

As has been reported in previous years, the data on host plant resistance were quite extensive and too voluminous to present in tables within this report. In addition, a rather wide range of insect preference tolerance, and/or resistance exists among the various entries and this varies from year to year based upon local environmental conditions and insect pest pressure. However, several entries that have been evaluated over several years show excellent promise for incorporation into an IPM program. Two entries, Robut 33-1 X NC 2214 and J11 X (M13 X NC 3083) have consistently demonstrated reduced insect damage. Several other entries have also been consistent in the level of resistance and yield. Under severe insect pressure, even the most resistant varieties have suffered yield reductions. The level of resistance available does, however, appear to offer an adequate level of protection under normal pest pressure. Dr. Ocampo's collaboration with Dr. Abilay has been highly beneficial. Overall pesticide use by the Institute of Plant Breeding has been reduced and Dr. Abilay's program has much better information on insect resistance. During the Peanut CRSP program, UPL Pn 10 and IPB Pn 2 were released and these have all been studied for insect susceptibility/resistance through this collaboration prior to release. Summary examples (Tables 1, 2 and 3) of screening evaluations indicate significant differences in the level of resistant available in entries under development and varieties already available.

Work in Quirino to evaluate the intercropping of peanut and banana has provided significant information over the past few years. Perhaps the most important component has been the dramatic differences in yield and insect performance in comparing trials at UPLB with the locations in the North. The farmers in this area still prefer BPI Pn 9 despite its known susceptibility to insects. While varieties like UPL Pn 6 performed well at UPLB, its performance lagged behind BPI Pn 9 at tests in Quirino. In addition, differences in the level of insect damage occurred in simulated shading to mimic intercropping practices (Table 4).

Dr. Cadapan continued work with *Trichogramma* sp. and with *Bacillus thuringiensis* in peanut with encouraging results. Biorational approaches on different peanut cultivars in the Zambales region have shown promising results for developing low risk peanut management. The major impact of Dr. Cadapan's work in this area has been recommended cultivar evaluation for local needs overlaid with conventional and IPM approach management strategies in both the wet (July-Oct 1994) and dry(Nov 1994- Feb 1995) seasons. These studies are focused primarily on the Lepidopterous pests found in peanuts. The use of released parasites and *B.t.* has proven very effective and acceptable in this region when using previously tested thresholds. In addition, these demonstration plots have been used to educate farmers concerning the use of IPM on peanuts. Table 5 shows summary data for one newly-released variety(UPL PN 10) under farmer (conventional) IPM systems.

ADDITIONAL STUDIES IN THE PHILIPPINES-Studies on the subterranean ant have been slow to develop partially due to the inherent difficulties associated with soil insect research Current knowledge indicates that poor crop stands are often directly related to this pest. Chemical control is the only option available for this pest and has been found to be effective. Previous studies by Drs. Cadapana and Ocampo have developed a comprehensive list of economic threshold and appropriate control responses. These are listed in Table 6.

THAILAND - Dr. Manochai Keerati-Kasikorn focused exclusively on the evaluation of peanut germplasm for resistance at Khon Kaen during the rainy season of 1994. This research included 10 lines from the International test-F6 lines, 22 lines from the International test ICRISAT germplasm, 22 lines from the Uniform

Nursery test #2, 12 lines from the International test-Insect Germplasm and 11 lines from the Uniform Insect Nursery. Tainan 9 and KK60-1 were used as check entries for standardized comparisons. Leaf damage by thrips and leafhoppers were evaluated at 38 and 64 days after planting and pod damage was evaluated at harvest. The results of these trials are summarized in Tables 7-11. In general, the thrips, leafhopper, and leafminer incidence was low at this location.

Dr. Turnjit Satayavirut also conducted germplasm screening trials at the Prabuddhabat Experiment Station. A total of 167 lines were evaluated including 33 lines from the F10 test, 50 lines from the IGT test, 37 lines from the ICRISAT test, and 40 line from the LHT test. This study was conducted during the rainy season from July to November 1994. Significant differences in insect abundance were observed and the lines with lower pest densities are listed in Table 12.

Sticky traps studies of sucking pests and their relationship to the incidence of viral diseases that they vector were continued at this site. Data collection is still underway and the results will be reported at a later date.

Drs. Satayavirut and Keerati-Kasikorn cooperated on several studies at Khon Kaen University. A study was completed during the rainy season that included visual rating of red mite damage on 40 selected lines of germplasm from NCSU. All other pests populations were low, but red mite densities were relatively high and varied significantly among the lines and warrants further research efforts.

A study using three cultivars (KK 60-3, Tainan 9, and SK 38) and 7 different insecticide treatment/management regimes in a split-plot design was conducted a Khon Kaen Field Crops Research Center. Population densities of thrips and leafhopper and aphid damage were monitored. While insect density was less on KK 60-3 its yield was significantly less and insecticides were valuable tools in preserving peanut yield from insect loss. Treatment schedules are included in Table 13 and thrips density and yield are found in Tables 14 and 15, respectively.

Recent efforts to initiate investigations into the effect of soil insects on peanut production in Thailand prompted a study at Prabudduhabat Experiment Station during 1994 to determine the effect of insect pod damage on the incidence of aflatoxin (*A. flavus*). This damage is caused by a pest complex including subterranean ant, termites, and white grubs. Results of this study indicate that more insect damage to seed is reflected in a higher incidence of aflatoxin (Table 16).

Since insecticides continue to play an important role in insect management in Thailand an efficacy trial was conducted at the Khon Kaen Field Crops Research Station during 1994. This study looked at several products on the insect complex and indicates numerous effective products are available as "rescue" treatments in an IPM program.

NORTH CAROLINA-Models for the lesser cornstalk borer and twospotted spider mite were validated during 1995. The lesser cornstalk model predicted no problems from this pest and none were observed despite drought conditions during July. The spider mite model predicted outbreaks in August which did occur and then forecast an abrupt population decline later that same month which was also confirmed with field observations. The integration of these models into an already existing system for peanut leafspot forecasting has greatly facilitated their implementation and adoption. County agents are using this information and relaying it directly to growers to reduce unnecessary pesticide use, refine current control strategies, and help target cultural practices to minimize the impact of these pest.

The ecological studies investigating the overwintering and dispersal of thrips into peanuts confirmed first year findings. Earlier studies have documented the migration into fields, but our understanding of their overwintering sites and mechanisms were much less clear. Studies during 1993-94 found that many thrips overwinter in the soil of fields that were in peanut the previous growing season and studies during the winter of 1994-95 confirmed these observations(Table 17). Thrips begin to emerge from the soil in mid April, about two week before most peanuts are planted and three weeks before peanut plants emerge. Studies for the winter of 1995-96 include an investigation of the ability of the tomato spotted wilt virus to overwinter in the thrips.

A study on the impact of tropospheric ozone on peanuts plants and twospotted spider mites was completed in the spring 0f 1995 and this work has enhanced our understanding of the factors that encourage outbreaks of this pest. Elevated levels of ozone, commonly observed in the peanut-growing areas of the Southeast, result in increased short-term ovipositional rates which results in a more rapid population increase(Table 18). The effect appears to be an indirect effect on the mite as it feeds on the ozone-injured peanut plant. These increasing levels of tropospheric ozone on a worldwide basis, would indicate the selection for more ozone tolerant cultivars as a possible mechanism for reducing mite outbreaks.

Research on biological control continues for foliage feeding pests. The use of "safer" pesticides to help reduce the potential for human and environmental risk is a critical issue for the U.S. peanut industry. Our goals in sustainable agriculture seek to minimize pesticide use, however,current technology limits our ability to effectively control all pests in this manner. At this time we can only seek to utilize those products best suited for sustainable programs.

Host plant resistance work in cooperation with Dr.Tom Isleib, peanut breeder at N. C. State University, evaluated thrips, leafhopper, and defoliator damage on 84 lines. These studies found significant differences in the level of damage among several entries. These data are still under final analysis and will be presented at a later date, but preliminary data for thrips injury are presented in Table 19. These studies do provide insight into the potential for integrating host-plant resistance more fully into sustainable production systems. The 1995 studies also revealed significant differences in the level of tomato spotted wilt virus among different lines. This is of great significance in light of the inability to directly manage this disease by controlling the vector, thrips. Data on thrips abundance and feeding injury are being compared to level of virus among plots and this analysis is still underway. Expanded studies on this area are planned in 1996.

Annual Plan of Work NCS/IM/TP - 1995-96

Rick L. Brandenburg
North Carolina State University

1. Research thrusts/endeavors planned

UNITED STATES

A. Enhanced understanding of thrips (Frankliniella fusca) and tomato spotted wilt the virus overwintering and the development of management strategiesthrough biological studies and management trials.

1. Studies to determine the overwintering of the thrips and the virus will be conducted throughout the peanut producing region and on the NCSU campus.

2. Key parameters used by thrips for host finding will be studied in early season trials by using varieties with different toleerances to thrips feeding and injury.

3. An improved understanding of the life history of thrips on Virginia-typepeanuts will be elucidated.

B. Improved pest management and development of sustainable systems through predictive modeling, trap cropping and cultural practices.

1. Use environmental monitoring and intuitive modelling to predict the presence of insect pests in peanuts and relate to IPM control strategies.

2. Conduct studies on biological control through preservation of endemic species.

3. Validate current models on lesser cornstalk borer and spider mites.

C. Host Plant Resistance

1. Expand effort in plant breeding program for resistance selection. Initial research in 1994 evaluated 84 entries for resistance to thrips, leafhopper and corn earworm damage.

2. Recruit Ph.D. student to examine resistance mechanisms in peanut. Evaluate the impact of environmental pollutants on host-plant suitability and the subsequent pest development.

PHILLIPINES

A. Wet season (July-October 1995)

1. Continue Germplasm Evaluation for Resistance against Peanut Insect Pest Complex. Coordinated evaluations of promising genotypes with Dr. R. Abilay. The genotypes evaluated will be based upon previous performance trials at UPLB and at outlying production areas.

2. Insect Pest Damage Assessment of Phillipines Seedboard Recommended Peanut Varieties at UPLB and at the Cagayan Valley Integrated Agricultural Research and Development Center.

Seven varieties, UPL Pn-2, Pn-4, Pn-6, Pn-8, BPI P9 and P2 and JL-24 will be sown and superimposed on this experiment is data collection for the PNUTGRO Model being evaluated by Engr. Andrew Buendia of PCARRD. Insect pest damage rating and insect counts will be done regularly throughout the crop's growth stage. Sticky and/or pheromone traps will be set up to monitor insect populations and their source around the area.

3. Continue the evaluation of biological control agents including *Trichogramma* and a newly-discovered *Erynia* fungal pathogen for utilization into agrower pest management program.

4. Expand host plant resistance work and pest management assessment trials into areas of Region II (Qirino and Isabela) and Pamganga province.

5. Determine the severity of damage caused by subterranean ants during the wet and dry season peanut croppings; describe the feeding behavior, colonization pattern and distribution of subterranean ants in the field; and evaluate the preference of the adult ants for the different feed materials including different major crop seeds.

B. Dry season (December 1995 - March 1996)

1. Completed Arthropod Diversity Studies in Peanut under Different Cultural Management Practices and Development of Sustainable Agricultural Systems.

Survey on peanut insect pest damage and population as well as on theirnatural enemies will be completed on different peanut cropping patterns (banana-peanut intercrop or corn-peanut intercrop) using the recommended Philippine Seedboard peanut varieties in Region II.

2. Documentation of IPM Benefit Through Insect Pest Management Packaging;, Field Valaiidation and Technology Transfer and Sustainable Agriculture Systems.

The tactics in the IPM package will be evaluated at a farmer's field in Region II and compared to the existing farmer's crop production practices. The package includes release of *Trichogramma* parasitoids, liming, use of microbial (B.t.) insecticides and other insecticides found effective in our screening trials at UPLB, based on ELT's. In addition, the use of trap crops will be evaluated.

3. Continued Germplasm Evaluation for Resistance against Peanut Insect Pest Complex.

The germplasm collection mentioned earlier will also be evaluated for insect pest resistance during the dry season at UPLB and/or Region II following the same procedures in the wet season trials.

4. Finalize evaluations of biological control agents for lepidopterous pest control. Agents include Trichogamma and a newly-discovered *Erynia* fungal pathogen for utilization into a grower pest management program.

5. Determine level of natural control in peanuts against major insect pests. Utilize large plot studies in various production regions to determine the potential and reliability of endemic biological control against major insect pests in the absence of pesticides.

6. Continued emphasis on crop loss assessment studies of major pests in Region II production area on improved cultivars.

THAILAND

A. Finalize Screening of Peanut Lines from North Carolina Collection for Insect Resistance. Prabudhabat Experiment Station, Khon Kaen Field Crop Research Center, and Kalasin Field Crop Research Center. Studies are focusing on those cultivars that have shown resistance in previous years and data will be reviewed and published for use on a regional basis.

B. Complete ecological and management studies on subterranean ants at RayongField Crop Research Center.

C. Insect Vector Studies - Migration and Alternate Hosts

1. Complete Studies on immigration of some insect vectors to peanut field by using sticky traps. A complete picture of vector movement as it relates to weather and crop phenology is being developed at Prabudhabat Exp. Station and Khon Kaen Field Crop Research Center. Correlation of thrips numbers and yellow spot virus and their effects on yield of selected peanut cultivars at Prabudhabat Exp. Station.

D. Publish results from the IPM studies on Tainan 9, KK60-3 (NC7) and Sukotai 38 (S.K. 38). Thesetrials will integrate cultural, biological, as well as chemical strategies at Khon Kaen Field Crop Research Center and farmers' fields in the N.E. part of Thailand and available for regional use.

E. Publish all research results and work with DOA and DOAE in the development of regional IPM training materials.

2. Training Planned

Two students will complete MS degrees in Entomology at UPLB funded by Peanut CRSP.

3. International Travel

Month:	December 1995
Destination:	ICRISAT, INDIA
Duration:	One week

Meet with collaborators and tour research plots. Conduct IPM workshopwith John Wightman and discuss future collaborative efforts between Peanut CRSP and ICRISAT in areas of sustainable agriculture. Possibly invite one or two HC scientists and scientists in Africa curently collaborating with ICRISAT.

4. Manuscripts, Presentations

Brandenburg, R. L. and B. M. Royals. 1994. Caterpillar control on peanuts, North Carolina, 1993. Insecticide and Acaricide Tests 19:93F.

Brandenburg, R. L. and B. M. Royals. 1994. Thrips control in peanuts, North Carolina, 1993. Insecticide and Acaricide Tests 19:94F.

Brandenburg, R. L. and B. M. Royals. 1994. Control of twospotted spider mite in peanuts, North Carolina, 1993. Insecticide and Acaricide Tests 19:95F.

Heagle, A. S., R. L. Brandenburg, J. C. Burns, and J. E. Miller. 1994 The Effect of Chronic Ozone and Carbon Dioxide on populations of Twospotted Spider Mite on White Clover and Peanut. J. Environ. Qual. 23:1168-1176.

Garcia, L., and R. L. Brandenburg. 1995. Spring movement of *Frankliniella fusca* into peanuts. In Thrips Biology and Management. B. L. Parker, M. Skinner, and T. Lewis (eds.) Proceedings of a NATO Advanced Research Workshop. Plenum Press. 636pps.

Funderburk, J. E., and R. L. Brandenburg. 1995. Management programs for Peanut Insects. p. 51-58. In Peanut Health Management. H. A. Melouk and F. M. Shokes (eds.) Amer. Phytopathological Soc. 117pps.

Brandenburg, R. L., D. A. Herbert, G. S. Sullivan, G.C.Naderman, and S. F. Wright. 1995. The Impact of Conservation Tillage on Pest Species in Peanuts. (under review).

Barbour, J. D. and R. L. Brandenburg, 1995. Impact of Turf and Timing of Corn Rootworm Treatments in Peanut on Beneficial Arthropods. J. Econ. Entomol. (30:447-462).

Hummel R., R. L. Brandenburg, and A. S. Heagle. 1995. The Effect of Chronic Ozone Exposures on Population Dynamics of Twospotted Spider Mite on White Clover and Peanut (in prep).

Hummel R., R. L. Brandenburg, and A. S. Heagle. 1995. The Effect of Ozone on Reproduction in the Twospotted Spider Mite (*Tetranychus urticae* Koch). Southeastern Branch meeting, Entomological Society of America, Charleston, SC

Table 1. Percent yellowing exhibited by UPL Pn-4 crosses. 1992-1994

IDENTITY	Wet Season 1992-93	Wet Season 1993	Dry Season 1994
UPL Pn 4 X NC 10247	8.67	0.1	28.0
UPL Pn 4 X GP-NC 343	6.96	0.1	10.7
UPL Pn 4 12.12	0.5	16.0	

Table 2. Insect damage and abundance on advance yield trial (AYT) entries, dry season. January to May 1994. UPLB.

Entry No.	Identity	Yellowing %	Defoliation %	Thrips Count	Aphid Rating (0-3)
1	IPB Pn 85-2-40	40.0abcde	3.8c	2.3a	0.7b
2	IPB Pn 85-3-86	252.6f	3.2bcd	2.7a	0.0d
3	IPB Pn 85-10-68	29.8ef	2.9bcd	2.0a	0.3c
4	IPB Pn 85-14-60	30.7def	3.2bcd	3.0a	0.3c
5	IPB Pn 87-26-4	46.6abc	3.9bc	1.3a	1.0a
6	IPB Pn 87-26-29	45.6abc	3.4bcd	3.0a	0.3c
7	IPB Pn 88-21-24	47.1a	4.3abc	2.3a	0.0d
8	IPB Pn 88-30-39	44.4abc	4.6ab	3.0a	0.0d
9	MM Pn 27	34.2cdef	5.8a	1.3a	0.3c
10	MM Pn29	42.5abcd	3.7bcd	2.3a	0.3c
11	MM Pn 29-1	33.8cdef	3.6bcd	1.7a	0.3c
12	UPL Pn4	32.0def	1.9d	4.7a	1.0a
13	Mahica	35.1bcdef	3.6bcd	1.7a	0.3c
14	Cina	30.7def	1.9d	4.7a	1.0a
15	ACC12	39.2abcde	2.8bcd	3.0a	0.3c
16	UPL Pn2	36.9abcde	3.34bcd	1.3a	0.3c
17	Khon Kaen 60-1	44.0abc	4.4ab	1.3a	0.3c
18	BPI Pn2	38.2abcde	3.4bcd	1.3a	0.3c
19	UPL Pn8	34.2cdef	2.9bcd	3.3a	0.3c

Means with the same letter are not significantly different at 5% level of significance.

Table 3. Percent yellowing exhibited by Philippine Seedboard peanut varieties. 1992-1994

IDENTITY	1993	1994
UPLB		
UPL Pn-2 0.45c	38.7a	
UPL Pn-4	0.55b	28.0abc
UPL Pn-6 0.59a	30.7abc	
UPL Pn-8 0.36d	18.7bcd	
UPL Pn-10	0.50bc	16.0cd
BPI Pn-9 (Check)	0.36d	22.7abc
Quirino Native (Check)	0.32d	

Table 4. Response of boiling type peanut trial entries on common field insect pests with varying shade level. January to May, 1994. UPLB.

Shade Level	Identity	Yellowing %	Defoliation %	Thrips Damage %
Exposed to Sunlight	IPB Pn 85-25	47.3bc	6.7bc	4.8a
	IPB Pn 85-35	46.7c	6.5bc	6.0a
	IPB Pn 85-45	46.0c	5.2c	4.8a
	IPB Pn 85-26	53.3a	5.8bc	5.0a
	IPB Pn 87-50	52.7ab	5.3bc	6.5a
	IPB Pn. 87-11	56.0a	6.3bc	6.2a
	IPB Pn 88-30	52.7ab	7.3b	7.0a
	IPB Pn 88-27	55.3a	9.3a	4.8a
	UPL Pn2	57.3a	9.7a	6.5a
One layer	IPB Pn 85-25	43.3abc	7.2a	5.0a
	IPB Pn 85-35	46.7ab	7.8a	7.0a
	IPB Pn 85-45	35.3c	7.8a	5.7a
	IPB Pn 85-26	46.7ab	6.8a	5.8a
	IPB Pn 85-50	40.0bc	7.5a	5.3a
	IPB Pn 85-11	50.0a	7.7a	7.3a
	IPB Pn 88-30	44.0abc	7.3a	5.7a
	IPB Pn 88-27	46.7ab	9.0a	6.0a
	UPL Pn2	41.3abc	9.0a	6.0a
Two layers	IPB Pn 85-25	4.8a	7.8a	6.7a
	IPB Pn 85-35	44.7ab	6.5a	6.0a
	IPB Pn 85-45	40.7ab	7.5a	5.7a
	IPB Pn 85-26	38.0b	6.5a	6.5a
	IPB Pn 87-50	41.3ab	8.3a	6.3a
	IPB Pn 87-11	40.7a	6.7a	6.2a
	IPB Pn 88-30	46.7ab	8.0a	5.7a
	IPB Pn 88-27	42.0ab	6.3a	6.2a
	UPL Pn2	43.3abc	8.3a	4.8a
Three layers	IPB Pn 85-25	32.7ab	4.7b	6.7a
	IPB Pn 85-35	34.7ab	6.3ab	8.0a
	IPB Pn 85-45	31.3b	6.3ab	6.0a
	IPB Pn 85-26	38.0ab	5.8ab	7.5a
	IPB Pn 85-50	33.3ab	7.8a	7.8a
	IPB Pn 87-11	34.7ab	5.8ab	6.8a
	IPB Pn 87-30	38.0ab	6.0ab	6.8a
	IPB Pn 88-27	36.0ab	6.0ab	6.3ab
	UPL Pn2	42.0a	8.3a	6.5a

Means with the same letter are not significantly different at 5% level of significance.

Table 5. Mean percent insect damage during first 12 weeks post plant UPL Pn10 peanuts under different management regimes. San Antonio, Zambales. July to October 1994.

	% leafhopper damage	% defoliation	% thrips damage
Farmer	28.48	24.94	3.37
IPM	19.71	14.15	2.32
Untreated	28.49	21.88	3.43

Table 6. List of the pest control measures used in the IPM Package.

ARTHROPOD	ETL	TREATMENT
A. Pest		
1. Aphids	when leaf begins to curl	handpicking/crashing
2. Thrips	40% leaflet bronzing	none
3. Leafhopper	50% leaflet yellowing 75% leaflet yellow	carbaryl carbaryl
4. Defoliators	40% defoliation 10% defoliation	Baciillus thuringiensis (Bactospeine)
5. Leafminer	50% damage early or late season 15% damage 60-90 DAP	none
6. Pod borers	3.85% pod damage (Furadan 3G)	carbofuran
B. Pest Complex		
1. Aphids Thrips Leafhopper	when any single pest reach ETL	carbaryl
2. Defoliators Leafminer	when any single pest reach ETL	B.t.-Bactospeine
3. Aphids Thrips Leafhopper Defoliators Leafminer	when any single pest reach ETL	monocrotophos

Table 7. Evaluation of Peanut Germplasm for Resistance to the Complex of Insect at Khon Kaen of 1994 Rainy Season

Entry No.	Identity	No. of damaged leaves/row						Pod Yield
		Thrips		Leafhopper		Leafminer		t/ha
		38	64	38	64	38	64	
1	NC7 x GP-NC 343	18.5	47.2	3.5	15.0	0.75	5.0	1.467
2	NC7 x NC 15745	26.2	41.2	5.25	12.7	0.75	8.25	0.949
3	NC6	11.0	33.7	2.25	8.75	0.75	6.00	0.754
4	Lampang x NC 15729	28.7	41.0	4.0	10.5	2.0	5.5	1.222
5	CES 101 x NC 342	31.5	55.2	4.0	13.0	0.75	9.5	0.837
6	CES 101 x NC 302	25.2	39.2	3.0	10.5	0.25	5.25	0.658
7	UPLPn4 x NC 10247	43.5	108	1.5	5.25	0.25	6.5	0.730
8	Tainan 9	38.5	65.7	3.25	11.7	0.00	6.25	1.238
9	KK 60-1	32.0	70.7	9.0	12.0	1.0	6.25	1.209
10	Tainan 9	49.2	67.5	12.7	9.0	0.25	6.265	1.351

Table 8. Leaf damage and yield of 22 peanut lines (International Test ICRISAT germplasm) evaluated for resistance to an insect complex at Khon Kaen University, 1994 rainy season.

Entry No.	Identity	No. of damaged leaves/row						Pod Yield
		Thrips		Leafhopper		Leafminer		t/ha
		38	64	38	64	38	64	
1	[Robut33-1xNC2821]x NC2232	21.2	108	1.5	15.5	0.25	8.5	1.046
2	Robut33-1xGP-NC343	10.0	33.5	3.0	31.0	0.25	4.75	1.488
3	Robut33-1xNC2214	7.0	49.2	1.5	11.0	0.25	11.7	0.775
4	Robut33-1xNC2214	11.7	37.7	1.0	9.0	1.75	18.2	1.127
5	M13xNC2214	23.0	39.0	7.0	18.7	0.75	8.5	0.464
6	M13xNC2214	29.5	55.0	10.2	13.7	1.0	12.0	0.928
7	Manfredi 68 XGP-NC343	24.0	63.52	4.25	42.2	0.5	5.75	1.220
8	Mani Pintara x [Robut 33-2xNC Ac2232	11.5	33.2	2.0	18.5	0.5	13.0	1.447
9	Mani Pintara x 33-1x NC Ac2232	18.2	28.7	2.0	17.2	0.5	15.0	1.304
10	TMV-10-NCAc2232	8.25	69.5	2.5	35.2	0.75	11.2	0.709
11	Manfredi 68xGN-NC343	21.2	98.0	4.0	26.0	0.0	17.2	1.184
12	Manfredi 69xGN-N C343	34.2	55.7	2.0	22.5	0.25	6.0	1.196
13	Tifspan x NCAc2232	20.0	26.2	3.0	27.0	0.25	9.5	1.454
14	Shulamit [Robut33-1x NC2214]	8.0	61.0	2.0	36.2	0.0	7.0	1.286
15	NC1107 x [NC2232 x NC2214]	31.7	83.5	2.75	14.0	0.0	12.0	0.784
16	NC2232 x NC2232 x NC2214]	26.2	79.2	1.5	17.0	0.25	12.2	1.071
17	[NC2232 x NC2214] x TG-17	19.2	78.0	1.75	26.5	0.25	7.75	1.417
18	[Goldin-1 x Faizpur 1-5] NC2232	25.2	102	3.0	19.0	0.0	6.75	2.035
19	[Goldin-1 x Faizpur 1-5] NC2232	30.0	66.5	2.5	19.2	0.0	13.5	1.474
20	GP-NC343	12.0	25.0	2.25	2.25	0.5	9.52	0.754
21	KK 60-1	50.0	70.0	10.0	45.0	0.252	7.25	1.018
22	Tainan 9	66.0	66.0	9.0	18.2	0.0	5.75	1.118

Table 9. Leaf damage and yield of 22 peanut lines (Uniform Insect Resistance Nursery Test #2) evaluated for resistance to an insect complex at Khon Kaen University, 1994 rainy season.

Entry No.	Identity	No. of damaged leaves/row Thrips 38	Thrips 65	Leafhnopper 38	Leafhnopper 64	Leafminer 38	Leafminer 64	Pod yield t/ha
1	NC7xGP-NC 343	19.0	45.0	7.5	37.0	0.7	8.2	1.417
2	"	37.5	32.0	9.7	34.5	0.0	11.2	1.680
3	"	33.2	55.2	8.0	36.5	0.0	12.5	1.094
4	"	31.5	41.2	5.0	24.5	1.5	15.5	1.021
5	"	55.7	57.0	4.5	39.0	0.2	14.2	1.620
6	"	28.2	37.0	10.0	42.4	1.2	12.0	1.533
7	"	59.5	38.5	5.7	31.7	0.7	8.0	0.841
8	"	33.0	32.7	8.7	33.0	0.2	11.0	1.263
9	NC7xNC 302	61.7	59.0	7.5	35.0	0.2	7.5	1.478
10	NC7x C 302	66.2	61.5	8.2	35.5	0.2	9.5	1.084
11	"	61.0	58.5	6.7	44.7	1.0	10.7	2.108
12	" (Susceptible check)	67.2	54.2	6.5	29.5	0.7	6.5	1.575
13	NC7xNC15745	34.0	52.0	9.5	25.7	0.7	7.0	0.871
14	NC7xNC15745	49.5	61.7	6.7	37.2	0.2	11.5	1.237
15	"	40.5	38.7	5.0	30.5	1.0	13.2	0.840
16	"	44.0	38.7	8.5	40.2	0.0	10.5	1.536
17	"	33.0	30.0	5.2	42.7	1.2	14.0	1.248
18	NC7xNC 10247	74.5	119.0	10.5	38.7	0.5	12.7	1.125
19	"	72.0	52.2	5.7	35.0	0.2	9.7	0.921
20	NC7xNC 301	45.0	50.7	7.2	37.7	0.7	7.2	1.194
21	KK 60-1	65.0	70.5	12.0	29.7	0.0	8.0	1.440
22	Tainan 9	84.2	67.7	12.0	28.0	0.0	6.2	1.110

Table 10 Leaf damage and yield of 12 peanut lines (International Test Insect germplasm) evaluated for resistance to an insect complex at Khon Kaen University, 1994 rainy season.

Entry no.	Identity	No. of damaged leaves/row Thrips 30	Thrips 45	Thrips 60	Leafhopper 30	Leafminer 45	Leafminer 60	Pod yield t/ha
1	NC6 x Early Bunch	25.5	45.7	5.7	27.7	0.7	7.5	1.219
2	3-P13-B2-P2-B2-B1-B1-B2	9.5	77.2	6.2	30.5	0.7	7.0	0.654
3	NC 5 x GP-NC 343	12.2	38.2	6.7	15.0	0.5	6.0	0.772
4	GP-NC 343 x NC 5	10.7	34.5	4.5	26.0	0.5	6.7	1.120
5	Bhairnwa Local	18.5	62.2	8.0	20.7	0.0	6.0	1.077
6	Comet	25.7	42.2	4.5	21.0	0.5	9.5	1.154
7	PI 459098, Toyokodachi	13.0	38.0	3.2	24.0	0.5	4.7	0.720
8	PI 467307, Ben-yang Shang-Shro Dur	15.7	41.7	7.2	23.7	0.2	6.2	1.058
9	NCAc17133RF	18.5	57.0	14.0	14.7	0.7	15.7	0.478
10	Florigiant	24.7	45.5	2.7	18.5	0.2	8.0	1.117
11	KK 60-1				14.0			
12	Tainan 9	25.5	76.7	11.5	24.7	0.5	5.2	1.417

Table 11 Leaf damage and yield of 11 peanut lines (Uniform Insect Nursery)evaluated for resistance to an insect complex at Khon Kaen University, 1994 rainy season.

Entry no.	Identity	No. of damaged leaves/row						Pod yield t/ha		
		Thrips		Leafhopper		Leafminer				
		38	64	38	64	38	64			
1	NC6	20.5	40.0	1.7	8.5	1.2	10.0	1.053		
2	GP-NC 343	13.7	38.5	3.0	8.5	1.2	9.5	0.860		
3	NC 302	21.0	46.0	4.5	7.2	1.0	7.0	0.775		
4	Robut 33-1 x NC2214	10.0	38.5	1.0	4.2	0.5	11.5	0.733		
5	"	3.5	19.0	1.2	6.7	1.0	15.2	0.641		
6	GP-NC 343 x NC 7	28.2	45.5	5.7	9.2	0.5	6.7	0.012		
7	GP-NC 343 x NC 5	21.2	34.5	2.7	11.5	0.7	5.2	0.665		
8	PI 467307	25.2	36.7	1.5	21.0	0.5	5.5	0.438		
9	NC 6 x NC 3033	14.7	29.2	1.5	8.2	1.0	6.0	0.642		
10	KK 60-1	28.7	66.2	11.2	8.7	1.0	4.5	1.087		
11	Tainan 9	33.2	69.5	12.0	12.7	1.2	5.5	1.131		

Table 12 Field screening of peanut germplasm from NCSU and local cultivars for resistance to insect complex. Entries with lowest insect pest abundance.

1994 Prabuddhabat Exp. Sta., Thailand.

F10-Test

NC7*GP-NC343, CES101*NC342, NC7, NC301, CES101*NC10272

IGT-Test

NC7, NC-AC 17133 RF, NC6, GP-NC343, NC6*NC3033

ICRISAT-Test

Robut 33-1*NC2214, Tifspan*NCAC 2232, Manipintar* (Robut 33-1* NCAC 2232), NC 1170*(NC 2232*NC 2214), NC6

LHT-Test

NC7*NC 10247, NC7*GP-NC 343, NC7*NC 10272, NC7*NC 15745

Table 13 Pest Management Treatment Schedules. Khon Kaen FCRS. 1994.

Treatment 1. insect control needed from plant emerging (E) to R2-stage (R2)
Treatment 2. insect control needed from R2-stage to harvesting stage (H)
Treatment 3. selective insecticide application for controlling only sucking insect pests.
Treatment 4. Treatment 1 incorporated with treatment 3.
Treatment 5. Treatment 2 incorporated with treatment 3.
Treatment 6. Treatment 1+2+3+Bt. (centari 30g/20 lits) application.
Treatment 7. Untreated (check)

Table 14. Thrips densities on KK-60-3, Tainan9 and SK38 peanut cultivars at different treatment applications, KK-FCRC, January-May 1994.

A. Cultivar	14 Feb	2 Mar	16 Mar	29 Mar	18 Apr	2 May
1. KK 60-3	1.1	2.3 a	12.3 a	21.1	3.2 a	1.8
2. Tainan9	2.0	11.4 b	22.9 b	29.0	5.3 b	1.5
3. SK 38	1.6	7.5 ab	18.9 ab	27.3	5.3 b	2.4
B. Treatment						
1. E-R2	1.7 ab	5.7	17.8	21.4	4.9	1.9
2. R2-H	1.2 a	6.7	19.0	31.2	3.1	2.1
3. Sucking Ins. Cont.	2.1 ab	9.5	15.0	28.6	5.6	1.1
4. T2+T3	0.6 a	8.1	21.1	24.2	5.5	2.9
5. T2+T3	4.0 b	7.5	18.7	24.6	4.3	1.3
6.T1+T2+T3+Bt	0.6 a	4.4	14.1	27.4	4.5	2.8
7. Check	0.7 a	7.5	20.5	23.1	4.3	1.2

Table 15. Yields (g/plot) of KK 60-3, Tainan9 and SK 38 peanut cultivars at different treatment applications, KK-FCRC, January-May, 1994.

	NOPL	GPOD	DPOD	POP	Pod wt. (g)	Seed wt. (g)	100 Seed wt. (g)
A. Cultivar							
1. KK 60-3	54.9 c	160.7 c	28.0	47.8	281.8 b	112.2 c	54.2
2. Tainan9	120.0 a	541.8 a	24.3	35.0	564.6 a	337.2 a	38.2
3. SK 38	82.4 b	286.9 b	21.3	29.5	440.7 a	191.8 b	36.7
B. Treatment							
1. E-R2	83.4	309.3	17.3	33.8	390.0	207.0 ab	43.0
2. R2-H	83.5	254.3	31.0	29.3	375.0	168.5 b	44.6
3. Sucking Ins. Cont.	90.2	359.3	28.2	37.7	413.3	200.1 b	42.1
4. T1+T3	86.9	358.7	21.9	40.3	465.0	212.1 ab	41.7
5. T2+T3	83.9	361.4	28.3	56.2	492.5	234.6 ab	47.5
6. T1+T2+T3+Bt	85.8	374.7	20.2	26.0	494.2	278.9 a	41.9
7. Check	86.6	291.0	25.1	38.8	373.3	195.0 b	40.4

NOPL - no. of plant/plot
GPOD - good pods/plot
DPOD - damaged pods/plot
POP - unfilled pod/plot

Table 16. % infection of *A. flavus*

Treatment	Seed	Pod
1) no damage	12.5	10.0
2) 1-25% pod damage	16.7	20.0
3) 26-50% pod damage	21.1	20.0
4) 1 hole (pod & seed)	63.0	52.0
5) 1 hold pod & 2 hole seed	45.8	32.0
6) 2 holes (pod & seed)	48.2	9.1

Table 17. OVERWINTERING THRIPS COLLECTIONS WITH PVC TUBES

N.C. Peanut Belt Research Station, Lewiston

	Winter 93-94	Winter 94-95				
	Peanut:	Peanut:	Corn:	Grass:	Woods:	Sticky Tube:
Dec.	2	3	12	0	0	1
Jan.	1	0	0	1	0	0
Feb.	0	0	1	1	0	2B*
1st Mar.	0	0	0	3	0	1
2nd Mar.	1	0	3	0	0	1
1st Apr.	20	24	5	7	0	12
2nd Apr.	101	80	17	85	0	57

*B = brachypterous

Note: 100 emergence tubes were set in peanut and in corn fields; 50 emergence tubes were set in grass and in woodborders; 8 sticky tubes were set (4 in the peanut field and 4 in the corn field).

Table 18. Mean cumulative number of eggs oviposited by first generation adult *T. urticae* feeding on two clones of *T. repens* exposed to a range of ozone concentrations.

Replicate	Clone	Mite age	Target ozone treatment level 15ppb	40 ppb	65 ppb	90 ppb	115 ppb
1	NC-R[a]	311[b]	0.5 ± 0.2[c]	0.3 ± 0.1	0.4 ± 0.2	3.4 ± 0.8	2.8 ± 0.4
		361	15.3 ± 1.0	12.2 ± 0.8	13.4 ± 1.2	21.5 ± 2.7	24.7 ± 1.6
		386	23.8 ± 1.6	19.1 ± 1.2	20.1 ± 1.9	31.2 ± 2.7	35.5 ± 1.9
		412	31.4 ± 2.5	28.7 ± 1.7	29.4 ± 2.5	38.3 ± 3.8	45.9 ± 2.3
	NC-S	311	0.0 ± 0.0	0.1 ± 0.1	0.5 ± 0.2	1.4 ± 0.3	2.0 ± 0.4
		361	8.9 ± 0.7	11.5 ± 1.2	17.6 ± 1.1	23.2 ± 1.4	23.8 ± 1.7
		386	16.5 ± 1.1	21.4 ± 2.1	28.7 ± 1.4	32.8 ± 1.6	33.0 ± 2.0
		412	24.8 ± 1.9	26.3 ± 2.4	40.4 ± 3.2	44.4 ± 3.2	45.4 ± 2.3
2	NC-R	286	1.1 ± 0.3	2.4 ± 0.3	3.4 ± 0.6	5.4 ± 0.9	5.6 ± 0.8
		313	8.3 ± 0.7	10.6 ± 0.5	12.8 ± 0.9	17.9 ± 1.3	17.5 ± 2.0
		336	17.9 ± 0.9	21.9 ± 0.8	23.8 ± 1.2	30.1 ± 1.1	31.5 ± 2.2
	NC-S	286	0.1 ± 0.1	0.1 ± 0.1	3.4 ± 0.4	4.9 ± 0.6	7.1 ± 0.6
		313	2.1 ± 0.5	6.2 ± 0.7	15.3 ± 0.8	19.0 ± 1.2	20.6 ± 1.2
		336	8.7 ± 0.9	15.5 ± 1.0	27.3 ± 0.9	33.0 ± 1.22	36.3 ± 1.2

[a]NC-R is a white clover clone resistant to ozone injury, NC-S is a white clover clone susceptible to ozone injury.

[b]Values represent the age of the first generation adult mite as measured in hours from original first generation egg production.

[c]Values are the means of 10 to 25 mites in two CSTR chambers ± SE.

Table 19.

95 L PIR: Preliminary Insect Resistance Test - Bertie Co., NC
Thrips Injury ratings. Scale-(0=low injury to 3=high injury) 1995

Entry	Identity of Parentage	Pedigree	Average 30 May 95	Average 15 June 95
1	NC 5/GPNC 343	Sel 01, F2-10-050-02:F10	2	2
2	GP-NC 343/NC 5	Sel 10, F2-05-128-02:F10	2.5	2
3	GP-NC 343/NC 5	Sel 11, F2-05-01-02:F10	2	1.5
4	GP-NC 343/NC 6	F2-S-S-B-10:F08	1.75	2
5	GP-NC 343/NC 6	F2-S-S-B-17:F08	1.75	1.25
6	NC-V 11/59-127,X85005-F1/3/ X85014-F1, NC-V 11 // (Sel 8), GP-NC 343/NC 5	F2-S-S-03:F07	2.25	1.5
7	NC-V 11/NC Ac 17090, X85006-F1 // X85012-F1, NC-V 11/GP-NC 343	F2-S-S-01:F07	3	2.75
8	NC-V 11/NC 8C, X85007-F1 // X85012-F1, NC-V 11/ GP-NC 343	F2-S-S-03:F07	2.25	1.75
9	NC-V 11 // Sel 10 (F2-05-01-01:F09), GP-NC 343/ NC 5	F2-S-S-09:F07	2.25	2.25
10	NC 7/NC 9, (F2-43-B-B-A04:F07) // (Sel 13), NC Ac 18229A/NC 2	F2-01-S-04:F06	2.75	2.25
11	NC 7/Florigiant, (F2-08-B-B-A03:F07) // N90021	F2-05-S-02:F06	1.75	1.5
12	GP-NC 343/NC 9	F2-B-02-01-02:F07	1.75	2.75
13	GP-NC 343/NC 9	F2-B-02-01-05:F07	1.75	2
14	GP-NC 343/NC 9	F2-B-02-01-07:F07	2.75	2.25
15	GP-NC 343/NC 9	F2-B-02-02-01:F07	1.75	1.75

Entry	Identity of Parentage	Pedigree	Average 30 May 95	Average 15 June 95
16	GP-NC 343/NC 9	F2-B-02-02-04:F07	1.25	2
17	GP-NC 343/NC 9	F2-B-02-02-06:F07	1.25	2
18	GP-NC 343/NC 9	F2-B-02-02-07:F07	1	2
19	GP-NC 343/NC 9	F2-B-02-02-09:F07	1.5	1.5
20	GP-NC 343/NC 9	F2-B-02-02-11:F07	1.75	2.5
21	GP-NC 343/NC 9	F2-B-02-03-02:F07	1.25	2.5
22	GP-NC 343/NC 9	F2-B-06-01-01:F07	2.25	2
23	GP-NC 343/NC 9	F2-B-09-05-01:F07	2	3
24	GP-NC 343/NC 9	F2-B-09-05-02:F07	3	2.25
25	GP-NC 343/NC 9	F2-B-09-05-04:F07	2.25	2.75
26	NC 7 // NC Ac 18473, NC Ac 17921/GP-NC 343	F2-02-03-01:F06	2.5	2.5
27	NC-V 11 // NC Ac 18473, NC Ac 17921/GP-NC 343	F2-02-01-05:F06	2.25	2
28	NC-V 11 // NC Ac 18473, NC Ac 17921/GP-NC 343	F2-02-02-05:F06	2.5	2.5
29	NC 9		2.5	2.25
30	NC-V 11 // Sel 11 (F2-05-01-02:F09), GP-NC 343/ NC 5	F2-01-02-01:F06	3	1.5
31	NC Ac 18423 // Sel 12 (F2-05-03-01:F09), GP-NC 343/NC 5	F2-01-03-02:F06	3	2
32	NC Ac 18423 // Sel 14 (F2-07-13-01:F09), GP-NC 343/NC 2	F2-03-01-01:F06	3	2.5
33	NC 9 // NC Ac 18472, NC Ac 17921/GP-NC 343	F2-01-02-02:F06	2.25	2.5
34	NC 10C		2.25	2
35	NC-V 11		2.5	2.25

Entry	Identity of Parentage	Pedigree	Average 30 May 95	Average 15 June 95
36	NC 9 // NC Ac 18473, NC Ac 17921/GP-NC 343	F2-03-01-02:F06	2	2.25
37	N90004/GP-NC 343	F2-S-S-01:F05	2.5	2
38	N90004/GP-NC 343	F2-S-S-02:F05	2.75	1.75
39	N90004/GP-NC 343	F2-S-S-03:F05	2	2
40	N90004/GP-NC 343	F2-S-S-04:F05	1.75	1.75
41	N90004/NC Ac 00302	F2-S-S-01:F05	2	2.5
42	N90004/NC Ac 00302	F2-S-S-02:F05	1.5	1.5
43	N90004/NC Ac 00302	F2-S-S-03:F05	1.75	2
44	N90004/NC Ac 00302	F2-S-S-04:F05	1.75	3
45	N90004/NC Ac 00302	F2-S-S-05:F05	1	1.75
46	N90004/NC Ac 00302	F2-S-S-06:F05	1.25	1.75
47	N90010E/GP-NC 343	F2-S-S-01:F05	1.75	2.5
48	N90010E/GP-NC 343	F2-S-S-02:F05	2	2.25
49	N90010E/NC Ac 00301	F2-S-S-01:F05	2	2.25
50	N90010E/NC Ac 00301	F2-S-S-02:F05	1.25	1.5
51	N90010E/NC Ac 00301	F2-S-S-03:F05	2.75	2.75
52	N90010E/NC Ac 00302	F2-S-S-01:F05	1.25	1.75
53	N90010E/NC Ac 00302	F2-S-S-02:F05	1.75	1.75
54	N90011/GP-NC 343	F2-S-S-01:F05	2	2.25
55	N90011/GP-NC 343	F2-S-S-02:F05	2.25	2.75
56	N90011/NC Ac 00301	F2-S-S-01:F05	1.75	2
57	N90011/NC Ac 00301	F2-S-S-02:F05	2	2.75

Entry	Identity of Parentage	Pedigree	Average 30 May 95	Average 15 June 95
58	N90017/NC Ac 00301	F2-S-S-01:F05	2.75	2.75
59	N90017/NC Ac 00301	F2-S-S-02:F05	2.5	2.5
60	NC 7/Florigiant, X84002 (F2-08-B-B-A01:F08) // NC Ac 00301	F2-S-S-01:F05	2.75	2
61	NC 7/Florigiant, X84002 (F2-08-B-B-A01:F08) // NC Ac 00301	F2-S-S-02:F05	2.5	2
62	NC 7/Florigiant, X84002 (F2-08-B-B-A01:F08) // NC Ac 00302	F2-S-S-01:F05	2	2
63	NC 7/Florigiant, X84002 (F2-08-B-B-A01:F08) // NC Ac 00302	F2-S-S-02:F05	2.25	1.5
64	PI 343365/PI 298845 ('Virginia Giant'), X87172 (F2-02-01:F05) // GP-NC 343	F2-S-S-01:F05	2.25	2.25
65	PI 343365/PI 298845 ('Virginia Giant'), X87172 (F2-02-01:F05) // NC Ac 00302	F2-S-S-01:F05	2	2.5
66	NC 6		1.5	2
67	NC 7		2.5	2.5
68	GP-NC 343		1.5	1.5
69	PI 121067 ('Indio from Argentina')		1.75	2.5
70	NC 7/NC 9	F2-14-B-B-A01:F08	2.5	2
71	NC 7/NC 9	F2-14-B-B-A04:F08	2	2.5
72	NC 7/NC 9	F2-16-B-B-A05:F08	2.25	2.75

GA/PV/N, TP

Peanut Viruses: Etiology Epidemiologyand Nature of Resistance

The Georgia Station, University of Georgia, USA
Institute for agricultural Research,
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INTRODUCTION

Groundnut (peanut) rosette (GR) is a major constraint in the production of peanut in Africa, and along with other viruses causes significant yield losses. In addition to actual yield losses, the periodic occurrences of rosette epidemics, causing total local crop loss, induces farmers to plant other more dependable crops. Before control measures can be implemented, the source of the rosette virus must be found, the nature of resistance elucidated, and resistance incorporated into drought-resistant, short season peanut.

Peanut stripe virus (PStV) was first discovered naturally infecting peanuts in the U.S. in 1982. After PStV was characterized and methods of identity developed, data from surveys in different countries of the world indicated that PStV is the most prevalent virus infecting peanut in S.E. Asia (Indonesia, Malaysia and the Philippines) and China. Recent reports documented the occurrence of PStV in India. Thus PStV is prevalent in areas where a large majority of peanut production occurs. Even though PStV may induce only low yield losses in the U.S.,data suggest much higher losses in S.E. Asia and China. The virus infects a sufficient number of plants to have an economic impact on total peanut production.

The incidence of tomato spotted wilt virus (TSWV) infecting peanut has increased in the U.S. in the past decade and has threatened production in a three county area of Texas. Increased incidence in the south eastern states has been noted in 1987, 1988, 1989, 1990, 1994 and more dramatically in 1995. The high yield loss by this virus poses a new constraint for peanut production.

The prominent viruses infecting peanut in Asia are different from those in Africa and the Western Hemisphere. Peanut bud necrosis virus (PBNV)(in the same group as TSWV) causes serious problems in India and Thailand. Peanut chlorotic streak virus (PCSV) has only been reported from India.

Peanut mottle virus (PMV) infects peanut on every continent where peanut is grown. Because it is seed and aphid transmitted, it infects more peanut plants on a global basis than any other virus.

This project was initiated in July 1990 and funds were available in Georgia about three months later. Memorandums of Understanding and Plans of Work were sent to all host countries but signed forms were not received in Georgia until 1991 so funds were not available to host country participants until spring 1991.

IMPACT

Resistance to GR in peanut lines that are acceptable to growers will permit greater production of peanut in Africa. This production will be by the subsistence farmers and provide a more reliable food crop that will enhance their diet. In general it will increase food production across the whole of Africa where peanuts are grown.

Identifying and transferring plant resistance to viruses in peanut and transforming peanut with virus coat protein genes are two systems that have the
greatest potential to reduce peanut virus diseases and thus provide a more sustainable crop which will increase the world food supply.

ACCOMPLISHMENTS

This project that started in 1990 had the main objective to transform (move foreign genes into peanut cells) and regenerate fertile plants from those transformed cells. The foreign genes that we are using are the genes from viruses that code for the production of virus coat protein. We have developed two methods that can move genes into peanut.

One method uses enzymes to digest immature cotyledonary tissue to remove the cell walls leaving individual cells without cell walls (called protoplasts). The protoplasts are placed in a solation with the virus coat protein genes and with electroporation (electric impulses) induce small openings in the protoplast. These protoplasts are stimulated to divide when placed in the presence of nurse culture cells. As these protoplasts divide, the cellular nucleic acid can incorporate the coat protein genes into the peanut nucleic acid. Using regeneration techniques, these transformed protoplasts are regenerated into whole fertile plants.

The second method used a bacteria to carry the foreign genes into peanut tissue. The virus coat protein genes are inserted into a binary vector which is incorporated into a strain of *Agrobacterium tumefaciens*. The bacteria is placed on immature leaf tissue which it can infect. After infection the bacteria are killed but they have already carried the foreign genes into the peanut cells. The tissues of peanut are then regenerated and we select the regenerates that have acquired the gene.

Currently we have plants growing in the greenhouse that have been transformed with the coat protein genes from TSWV, PStV and PCSV. These plants either have set seed or are currently setting seed and thus are fertile. All plants will be permitted to set seed and carried to the third generation where the progeny are expected to be homozygous for the transformed trait.

Additionally, we are selecting possible transformants that have the coat protein gene for the groundnut rosette assistor virus and bud necrosis virus.

A comprehensive package for virus control of peanut in Asia has been developed (S. Wongkaew - in press). The basic tenents of this package incorporates plant resistance, use of virus free seed and cultural practices such as adjustment of sowing date, using high plant density,and intercropping peanut with fast growing non-host crops.

In Africa, the program has relied on traditional breeding programs for the selection of traits to counteract the effects of virus infection. This program has been highly successful by releasing three varieties (M412.80I, MDR8-19 and K720.78) having resistance to groundnut rosette.

Thus assimilating the accomplishments in Asia, Africa and the U.S. with an additional three to five year research effort should provide sufficient control of virus diseases to lower the constraints of peanut production.

GOALS

The overall goal of this coordinated research program is to develop strategies and control measures to minimize incidence of the major peanut virus diseases, to foster interdisciplinary studies between countries and to develop linkages between supporting food production agencies. This will primarily be accomplished by strengthening peanut production programs in the host countries and the US through training and active research projects.

OBJECTIVES

Host Country - Nigeria

Phindile Olorunju completed her Ph.D. degree at the University of Georgia in virology/breeding. She has carried out a program of breeding for resistance to GR. Currently most African growers would like different

resistant lines (RMP-12 has a mottled seed coat, RG-1 is not proper size, etc). Other studies (objectives) were to get GR resistance into a range of season lengths, develop state trials and demonstration plots to assess disease and agronomic performance at a range of sites in Sudan and Guineas Savanna regions and to show farmers that resistant lines prevent epidemics of GR. Other objectives were to multiply seed of resistant released lines so they are readily available to farmers; interact with other Peanut CRSP programs such as the breeding project in Burkina Faso and the ICRISAT Sahelian Center for dissemination of ideas, information and breeding material.

Host Country - Thailand

Techniques for identification and manipulation of the peanut viruses have been established. As the virus surveys are completed, the major emphasis has been placed in various avenues that may lead to disease suppression or control. Some peanut cultivars have been identified with low rates of seed transmission to PStV and PMV. Studies were directed to determine if this trait is genetically controlled, and if so, to work with plant breeders to incorporate this resistance into acceptable peanut cultivars. A US peanut cultivar, Southern Runner, has resistance to TSWV. This was tested for resistance to BN and yellow spot, diseases of peanut in S.E. Asia caused by an apparent relative of TSWV. As resistant lines or transformed peanut plants become available from the US project they will be tested for effectiveness in S.E. Asia.

United States

Studies are being completed on the nature of the TSWV epidemics in peanut such as effects on yield, thrips as vectors, virus spread and field screening of commercial cultivars for resistance. This research project will also involve the transformation of peanut (Arachis hypogaea) with the DNA sequence coding for the coat protein of several peanut viruses to which a GUS marker gene will be attached.

Organization/Personnel:

A.	**U.S. Lead Institution:**	University of Georgia (UGA)
	Principal Investigator	Dr. James W. Demski Georgia Experiment Station, Griffin, GA 30223
	Co-Principal Investigator	Dr. Mike Deom Plant Pathology University of Georgia Athens, GA 30602
B.	**Nigerian Counterpart Inst:**	Institute of Agricultural Research (IAR) Ahmadu Bello University, Samaru, PMB 1044 Nigeria
	Principal Investigator:	Dr. Phindile Olorunju
	Collaborative Investigator	Dr. Steve Misari
C.	**Thailand Counterpart Inst:**	Faculty of Agriculture, Khon Kaen University, Khon Kaen 40002, Thailand
	Principal Investigator:	Dr. Sopone Wongkaew
	Collaborative Investigator:	Dr. Duangchai Choopanya
D.	**Informal Cooperation:**	ICRISAT, Patancheru P.O., A.P., 502324, India
	Principal Investigator:	Dr. D.V.R. Reddy

APPROACH

Host Country - Nigeria

1. Inheritance of resistance studies were conducted in a diallel test with eight peanut genotypes (28 crosses). The studies were designed to determine the nature of resistance to GR, the number of host genes involved in resistance, individual early-maturing F_3 plants with resistance to GR, and F_2 and F_3 plants with acceptable agronomic characteristics. Selection of F_2 and F_3 plants should provide the basis for this breeding program which seeks improvement in yield performance and incorporation of new traits such as drought resistance associated with resistance to GR. Screening for resistance to GR in all breeding generations includes both field and laboratory/screenhouse studies. Field screening was done by providing virus inoculum pressure by planting aphid-infected plants in alternate rows with test genotypes. The field inoculum included both GRV and GRAV. Mechanical inoculation with GRV only was done in the laboratory/screenhouse tests. The two methods are useful in identifying interactions between the two viruses that might affect certain agronomic characteristics. Principal investigators (host country and U.S.) maintained communication with Peanut CRSP personnel having breeding programs (Tx/BCP/WA) in other W. Africa countries. Site visits to the breeding plots for selection of virus resistance were made when appropriate.

2. Resistant (example-RMP-12) and susceptible (example-MK374) peanut lines were increased and given to farmers in diversified areas. These seed were planted as demonstration plots to convince growers of the value of using resistant lines to prevent GR epidemics.

Host Country - Thailand:

3. More emphasis was placed on BN since it has been identified as the prominent virus disease in Thailand. These studies included improved diagnostic studies, identification of vectors, virus isolate characterization and epidemiological studies.

4. Since PStV is so important in other S.E. Asia countries, a geographical area with no PStV incidence the previous year was selected. Plots of approx. 10 m^2 with 20 m spacing between plots were established. Maize was grown as a barrier crop between plots. Spacing within and between rows was approx. 30 cm. Randomly spaced plants (100 in each plot) were inoculated at the 1st true leaf stage with PStV. The infected plants were tagged. Yield data from the mechanically infected plants and the same number of healthy plants were compared in each plot.

5. To determine virus incidence, a single green leaf was taken from each of 10 plants per acre, at random throughout the field. Incidence per county, state or specified areas was obtained by dividing the total number of plants infected by the total number of plants sampled. To detect other viruses, additional samples were taken from plants with virus like symptoms.

6. Seed from the U.S. Plant Introduction Unit or the ICRISAT collection were screened for virus resistance in greenhouse and field tests. Field tests were conducted with infected source plants placed in the edge rows and internal plot rows separating test plants. Greenhouse tests were by mechanical inoculation. Non-seed transmission of PMV has been identified in some peanut lines and these lines were tested for non-seed transmissibility with PStV.

United States:

7. Arachis hypogaea types and 'wild' peanut types were acquired from the U.S. Plant Introduction Station. Seedlings, rooted cuttings and other progeny were mechanically and vector inoculated (separately with TSWV, PStV and PMV) using general virology procedures. Lines showing promise of resistance were field planted and subjected to inoculum pressure from infected source plants. Interspecific lines were also tested by mechanical and vector inoculation.

8. Primary source of virus was sought by serological tests of weeds and other plants growing in areas where TSWV epidemics have occurred in prior years. Thrips vectors were trapped as they emerge from the soil and serologically tested for TSWV. Plots were devised to test for internal virus spread as opposed to continual virus source from outside the plots. TSWV infected peanut seedlings were transplanted into some plots but not others and comparative incidence in each plot analyzed. Known TSWV source plants (peanuts, datura, tomato or tobacco) were placed in and near healthy peanuts and spread was carefully monitored.

9. Four approaches for transformation were attempted. The first approach involved electroporation of peanut protoplasts in a solution containing the coat protein sequence for peanut stripe virus. Electroporated protoplasts were plated onto a GUS selection medium and those protoplasts showing incorporation of the coat protein sequence into their genomes, as determined by the GUS assay, were regenerated into complete plants. The second approach involved the direct bombardment of cultured peanut shoot tips or meristems utilizing the particle gun. In this procedure the coat protein sequence is bound to microscopic tungsten or gold particles that are forcibly "shot" into the cells or tissues. Incorporation of the coat protein sequence into the peanut genome is determined by the GUS assay. The third approach placed the coat protein gene in solution with protoplasts and polyethlene glycol to induce protoplast penetration by this gene. The fourth used Agrobacterium to carry the genes into the peanut tissue.

ACCOMPLISHMENTS IN DETAIL

Africa

Emphasis has been concentrated on development of agronomically acceptable rosette resistant peanut cultivars adapted to the different ecological zones. Available genetic resistance in cultivated peanut has been demonstrated in a limited range of germplasm. Most of these are late maturing Virgina types. Select crosses have been made at the Institute for Agricultural Research (Nigeria) between resistant and susceptible genotypes. Recovery of resistant plants was low. Resistant lines that matured within 110 days were selected and advanced through a modified bulk breeding method. Selected lines were entered into replicated yield trails where other agronomic characteristics were monitored (Table 1). The resistant best yielders were tested in 1995 (data being assembled).

Cooperative studies with ICRISAT to obtain early maturing resistant varieties used a short season genotype ICGM 751/754 (purified KH 241 D) in crosses with sequentially branching and alternately branching parents. Single plant selections from the rosette disease nursery in Malawi and advanced to the F_4 as progeny plant bulk were tested in Nigeria. These results are given in Table 2.

Asia

Of the 26 viruses known to naturally infest peanut, PBNV, PStV, peanut clump virus (PCV), and cucumber mosaic virus (CMV) are the most important in the Asia-Pacific region. Based on research to date, the current control measures have been prescribed.

Peanut bud necrosis - Five methods using cultural practices appear to be effective. These are: adjustment of the sowing date, using high plant density, intercropping peanut with fast growing non-host crops, eradication of alternate hosts and using plastic mulch. In India it is advised to sow peanut early to avoid the thrip migratory flights so the plants are sufficiently developed to withstand infection by PBNV. Such effect has also been observed in Thailand. Peanut sown earlier in December or later in February appear to have lower PBNV incidence than those sown in mid January. Avoid planting peanut in the dry season is a good alternative. In Thailand PBNV seems to be prevalent only during the dry season.

Using high plant density is also effective in reducing losses from PBNV. This is due mainly to the compensation effect of the surviving plants. But dense plant canopy is known to reduce landing of thrips hence spreading of the disease. Canopy closure in the shortest time is less attractive to the vectors. Farmers are advised to sow good

quality seed that has been treated to avoid stand thinning by pests and diseases. It was found that the incidence of PBNV in crops that were sown at low densities was proportionately higher than in those sown at high densities.

Intercropping peanut with a fast growing cereal, such as corn, millet or sorghum reduces the incidence of BND. In thailand apart from corn, peanut is often intercropped with sugarcane which is also effective in reducing PBNV incidence. Attempts have been made to screen for peanut lines resistant to PBNV. Despite intensive efforts over many years by ICRISAT, none of the *Arachis hypogaea* genotypes tested has proved to be totally PBNV resistant. However, lines with a high level of field resistance as well as a good agronomic characteristics have been identified. Among them are ICGVs 91056,90013,90009,86598,91183,91228,86325, and 91229. Some of these lines such as ICGV 86388 performs well across India. This line also has lower PBNV incidence when tested in Thailand. Its lower infection rate when inoculated with the virus indicates its possible resistance to PBNV infection. It is expected that within a few years there should be a released cultivar that has both high yield and durable BND resistance.

Peanut stripe virus - Although PStV is detected in many weed species in peanut fields its source of primary inoculum appears to be mainly from infected peanut seed. The transmission rate of 2-5% is sufficient to lead to an epidemic of 100% incidence at harvest because of the subsequent dissemination by aphids. Therefore production and distribution of virus-free seed should be the method of choice in controlling the disease. In areas where farmers keep their own seed stocks, which is a common practice in most Asian countries, they should be advised to collect seed only from healthy-looking plants. Preparing a separate seed plot is also a good alternative, where intensive care is given to eliminate all infected plants. In some countries where government sectors play a key role in seed production and distribution, consideration should be taken in finding locations, cropping systems or seasons that are disease-free to produce seed stock. International exchanging of germplasm should take extreme precaution especially from countries where PStV is prevalent. It may be necessary to restrict the export of seed from infested areas. Plant quarantine units should develop expertise and facilities to detect PStV in seeds. At present, there is a nondestructive ELISA protocol that can be used for testing peanut seed. Like any other disease, cultivation of resistant cultivars should be the most effective method in PStV management, but resistance to PStV has not yet been found in cultivated peanut. The screening of approximately 10,000 accessions from ICRISAT's germplasm bank, in Indonesia yielded only PStV tolerant lines. However, resistance to PStV has been identified in some wild *Arachis* species and attempts should be made to transfer this resistance to *A. hypogaea*.

Another approach is to develop peanut cultivars with very low or no seed transmission. This approach has proved to be successful in controlling PMV. This strategy would reduce both sources of primary inoculum and subsequent spread by aphid vectors. Promising genotypes have been identified in China and in Thailand. In China 7 genotypes have been screened out from over 1300 entries tested. These genotypes showed less than 1% PStV seed transmission each in three consecutive tests.

In general, insecticides have not been effective in controlling the virus vectors of non-persistently transmitted viruses such as PStV. One method of virus reduction has been the use of silver-colored plastic film. Because of its high cost, it may not be practical in routine peanut production but may have merit in virus-free seed production.

United States

Using the enzymes cellulase, macerozyme, and/or hemicellulase, tissues of young peanut leaflets or immature cotyledons were digested to obtain protoplasts.

An electroporation medium containing 50 mM glycine or 10 mM glycylglycine (glygly), 70 mM potassium glutamate, and 0.4 M mannitol was evaluated for its ability to improve transient β-glucuronidase (GUS) expression in immature cotyledonary protoplasts of peanuts(Table 3). GUS activity in electroporated protoplasts was 8- to 430-fold greater than that obtained using any of four other commonly employed poration media. Analysis of viability and histochemical staining of protoplasts indicated that electroporation using the glycine- or glygly-based poration medium resulted in increased protoplast viability and GUS expression when compared with other poration media. Replacement of glygly with MES or HEPES buffers significantly reduced the level of GUS expresssion electroporated protoplasts. Up to 10% of the protoplasts divided and formed compact callus colonies.

The protoplast plating efficiency was correlated with both the length of the nurse cell co-cultivation period and the protoplast plating density. The optimal nurse culture duration was 14d. The optimal plating density was 2 X 10^4 protoplasts/ml plating medium. Multiple shoots (up to 10 shoots per colony) were readily regenerated from protoplast-derived callus after transfer of callus to semi-solid medified MS medium containing 0.5 mg l^{-1} NNA and 1 mg l^{-1} BA. Plantlets with normal leaflets were obtained by rooting shoots on porous rootcubes saturated with modified MS medium containing 1 mg l^{-1} NAA (Table 4). Using electroporation the TSWV coat protein gene has been inserted into peanut protoplasts and these protoplasts regenerated into fertile plants that are growing in the greenhouse.

Fertile transgenic plants of peanut cv. New Mexico Valencia A were produced using an *Agrobacterium*-mediated transformation system. Leaf section explants from *in vitro* grown seedlings were inoculated with *Agrobacterium tumefaciens* strain EHA105 harboring the binary vector pB1121 containing the genes for β-glucuronidase (GUS) and neomycin phosphotransferase II (NPTII). Inoculated leaf explants were cultured on shoot regeneration medium containing kanamycin. Putative transgenic shoots were identified by screening for GUS activity and the expression of NPT II. Approximately 10% of the shoots regenerated on selection medium were GUS-positive. Five independent lines of fertile transgenic peanut plants were obtained; a frequency of 0.3% of the leaf explants inoculated. On average, 240 d were required between seed germination for explant preparation and the production of mature T_1 seed by T_0 plants. Molecular analysis of T_0 and T_1 transgenic plants confirmed the stable integration of the foregin genes into the plant genome. The GUS gene segregated in a 3:1 Mendelian ratio in the T_1 generation (Table 5). Using this same protocol, the coat protein gene for PStV and PCSV has been inserted into peanut leaf tissue and the transformed cells regenerated into fertile plants. Plants having these genes are currently being propogated in the greenhouse.

The complete nucleotide sequence of the S RNA of peanut bud necrosis virus (PBNV) has been determined. It has 3057 bases, contains inverted repeats and has an ambisenese coding strategy. The sequence contains two open reading frames (ORFs), one of 1320 nucleotides located on the viral RNA (positive sense) that encodes for a Mr 49.5 kDa protein, and 831 nucleotides on the complementary strand (negative sense) that encodes for a Mr 30.6 kDa protein. ORF coding for the 30.6 kDa protein was expressed in *E. coli* utilizing pET-15b vector and immuno-blot analysis of expressed protein with polyclonal antisera against N protein confirmed that it is the N protein of PBNV. Amino acid sequence comparison of N and NSs proteins revealed identities of 22-34% with the reported tospovirus isolates of serogroups I, II, and III, whereas it had 82-86% identity with viruses in serogroup IV, watermelon silver mottle virus (WSMV) and tomato isolate of peanut bud necrosis virus (PBNV-To). From this data it is apparent that PBNV belongs to a highly diverged group that includes WSMV and PBNV-To. Two subgenomic RNA species detected in PBNV infected tissue corresponded to the predicted sizes (1.65 and 1.4 kb) of the NSs and N mRNAs. The absence of significant homology between PBNV and other tospovirus members of serogroups I, II, III and high degree of homology with WSMV and PBNV-T0 suggest that PBNV should be included in serogroup IV.

Using the coat protein gene from PBNV and the coat protein gene from the groundnut rosette assistor virus (obtained from the Scottish Crops Research Institute), we have initiated the transformation of peanut using *Agrobacterium*. These are currently in the selection process and hopefully we will have fertile transformants in the near future.

PUBLICATIONS AND PRESENTATIONS:

Reddy, D.V.R., J. A. Wightman, R. J. Beshear, B. Highland, M. Black, S. L. Divivedi, J. W. Demski, D. McDonald and D. H. Smith. 1991. Bud necrosis-a disease of groundnut caused by tomato spotted wilt virus. ICRISAT Information Bulletin. No. 31. 20 pp.

Sreenivasulu, P., J. W. Demski, D.V.R. Reddy, R. A. Naidu, and A. S. Ratna. 1991. Purification and serological relationships of tomato spotted wilt virus isolates occurring on peanut (*Arachis hypogaea*) in the USA. Plant Pathology 40:503-507.

Olorunju, P. E., C. W. Kuhn, J. W. Demski, S. M. Misari, and O. A. Ansa. 1991. Disease reactions and yield performance of peanut genotypes under groundnut rosette and rosette free field environments. Plant Dis. 75:1269-1273.

Culbreath, A. K., A. S. Csinos, T. B. Brenneman, J. W. Demski, and J. W. Todd. 1991. Association of tomato spotted wilt virus with general chlorosis and root necrosis in peanut. Phytopathology 81:810-811.

Sreenivasulu, P., C. W. Kuhn, R. A. Naidu, J. W. Demski, D.V.R. Reddy, and M. V. Nayudu. 1991. Viruses infecting peanut (*Arachis hypogaea*): Taxonomy, identification and disease management. Univ. of Georgia, Research Bulletin 406, 34 pp.

Demski, J. W., R. Pittman, J. Todd, and A. Culbreath. 1991. Identification and field resistance to tomato spotted wilt virus in groundnuts. International Arachis Newsletter 10:23-24.

Demski, J. W., R. N. Pittman, J. W. Todd and A. K. Culbreath. 1991. Identifying resistance to viruses in peanuts. In the 1991 Georgia Peanut Research - Extension Report 2:131-132.

Culbreath, A. K., J. W. Todd, and J. W. Demski. 1991. Influence of tomato spotted wilt virus on yield of florunner peanut. Phytopathology 81:1136. (abstr.)

Porter, D. M., J. W. Demski and P. M. Phillips. 1991. First report of tomato spotted wilt virus in Virginia. Plant Disease 75:431.

Culbreath, A. K., A. S. Csinos, T. B. Brenneman, J. W. Demski and J. W. Todd. 1991. Association of TSWV and foliar chlorosis in peanut in Georgia. Plant Disease 75:863.

Olorunju, P. E., C. W. Kuhn, J. W. Demski, S. M. Misari, and O. A. Ansa. 1992. Inheritance of resistance in peanut to mixed infections of groundnut rosette virus (GRV) and groundnut rosette assistor virus and a single infection of GRV. Plant Dis. 76:95-100.

Li, Z., J. W. Demski, R. L. Jarret, R. N. Pittman, and K. B. Dunbar. 1992. Plant regeneration from protoplasts of *Arachis paraquariensis* Chod. et Hassl. nurse culture method. Am. Soc. Plant Physiologists (abstr.) Aug. 1-5 in Pittsburgh, PA.

Li, Z., J. W. Demski, R. L. Jarret, R. N. Pittman, and K. B. Dunbar. 1992. Transformation of peanut (*Arachis* spp.) with a peanut stripe virus coat protein (PStV-CP) gene via particle bombardment and polyethylene glycol (PEG) treatment of protoplasts. Am. Peanut Res. and Ed. Soc. (abstr.) July 7-10 in Norfolk, VA.

Culbreath, A. K., J. W. Todd, and J. W. Demski. 1992. Comparison of hidden and apparent spotted wilt epidemics in peanut. Am. Peanut Res. and Ed. Soc. (abstr.) July 7-10 in Norfolk, VA.

Todd, J. W., J. R. Chamberlin, A. K. Culbreath, and J. W. Demski. 1992. Spotted Wilt Disease (TSWV) Incidence in peanut following various insecticide application regimes for thrips vector control. Am. Peanut Res. and Ed. Soc. (abstr.) July 7-10 in Norfolk, VA.

Li, Zhijian, J. W. Demski, R. L. Jarret, R. N. Pittman, and K. B. Dunbar. 1992. Protoplast regeneration and transformation of peanut (*Arachis paraguariensis* and *hypogaea*) with a peanut stripe virus coat protein gene (PStV-CP). ICRISAT, Patancheru, India (abstr.) 24-27 April 1992 in Wageningen, the Netherlands.

Chamberlin, J.R., J.W. Todd, A.K. Culbreath, W.C. Johnson and J.W. Demski. 1992. Post-harvest management of tobacco thrips (Thysanoptera:Thripidae) overwintering in peanut fields. J. Entomol. Science. 28:433-446.

Culbreath, A. K., J. W. Todd, J. W. Demski, and J. R. Chamberlin. 1992. Disease progress of spotted wilt in peanut cultivars Florunner and Southern Runner. Phytopathology 82:766-771.

Culbreath, A. K., J. W. Todd, and J. W. Demski. 1992. Productivity of Florunner peanut infected with tomato spotted wilt virus. Peanut Science 19:11-14.

Sreenivasulu, P., and J. W. Demski. 1992. Isolation and separation of common peanut potyviruses. Int.J.Trop. Plant Dis. 10:241-251.

Subrahmanyam, P., S. Wongkaew, D.V.R. Reddy, J. W. Demski, D. McDonald, and D. W. Smith. 1992. Field diagnosis of groundnut diseases. Information Bulletin No. 36. ICRISAT, Patancheru P. O., A. P. 502324 India 79 pp.

Chamberlin, J. R., J. W. Todd, R. J. Beshear, A. K. Culbreath, and J. W. Demski. 1992. Overwintering hosts and wing-form of thrips (*Frankliniella* spp.) in Georgia: Implications for management of spotted wilt disease. Environ. Entomol. 21:121-128.

Warwick, D.R.N. and J. W. Demski. 1992. Peanut stripe virus (PStV) transmission in peanut seeds. Fitopatol. Bras. 17:389-392.

Culbreath, A. K., J. W. Todd, J. W. Demski, B. D. Crowe and S. McKeown. 1992. Disease progress of tomato spotted wilt virus in Florunner peanut as indicated by serological assays and foliar symptoms. In the 1991 Georgia Peanut Research - Extension Report 2:136-137.

Sreenivasulu, P., J. W. Demski, C. W. Kuhn and R. G. Christie. 1992. Characterization of a necrosis strain of peanut stripe virus infecting beggarweed in Georgia, U.S.A. International Arachis Newsletter 11:21-23.

Demski, J. W., D.V.R. Reddy, S. Wongkaew, Z.Y. Xu, C. W. Kuhn, B. G. Cassidy, D. D. Shukla, N. Saleh, P. Sreenivasulu, R.D.V.J. Prasada Rao, and D. McDonald. 1993. Peanut stripe virus. ICRISAT Information Bulletin No. 36. 16 pp.

Culbreath, A. K., J. W. Todd, J. R. Chamberlin and J. W. Demski. 1993. Immunodetection of asymptomatic and symptomatic tomato spotted wilt virus infection of peanut: temporal dynamics. In the 1992 Georgia Peanut Research - Extension Report. pp. 173-175.

Culbreath, A.k., J.W. Todd, D.W. Gorbet and J.W. Demski. 1993. Spotted wilt apparent disease progress in the component lines of southern runner cultivar. Peanut Science 20:81-84.

Ribeiro, G. P., R.D.V.J. Prasada Rao, R. Pittman, D.V.R. Reddy, K. Dunbar and J. W. Demski. 1993. In vitro inoculation and maintenance of peanut viruses. Presented at the XXVI Congresso Brasleiro De Fitopatologia, Aracaju, S.E. Brazil on 16-21 De Agosto 1993.

Li, Z., J. W. Demski, R. L. Jarret and R. N. Pittman. 1993. Development of transformation systems for wild and cultivated peanuts for the integration of virus coat protein genes. Presented at the Peanut Virus Disease Working Groups Meeting 15-19 August 1993 in Dundee, Scotland. ICRISAT, Patancheru, 502324, India.

Demski, J. W. and M. Deom. 1993. The peanut CRSP virus project. Presented at the Groundnut Virus Disease Working Groups Meeting 15-19 August 1993 in Dundee, Scotland. ICRISAT, Patancheru, 502324, India.

Li, Z., R. L. Jarret and J. W. Demski. 1993. Plant regeneration from protoplasts of *Arachis* spp. In Plant Protoplasts and Genetic Engineering. Y.P.S. Bajaj, editor, Springer verlag, Berlin, New York. (In press)

Todd, J. W., J. R. Chamberlin, A. K. Culbreath and J. W. Demski. 1993. Timing and duration of vector management in relation to spotted wilt disease incidence in peanut. Presented at American Peanut Research and Education Society meetings, 13-16 July 1993 in Huntsville, Alabama.

Prasada Rao, R.D.V.J., G. P. Ribeiro, R. Pittman, D.V.R. Reddy and J. W. Demski. 1993. Reaction of *Arachis* germplasm to peanut stripe, peanut mottle and tomato spotted wilt virus. Peanut Science 20:115-118.

Li, Z., R. L. Jarret, R. N. Pittman, K. B. Dunbar and J. W. Demski. 1993. Efficient plant regeneration from protoplast of *Arachis paraguariensis* chod. Et Hassl using a nurse culture method. Plant Cell Tissue and Organ Culture 34:83-90.

Yang, Guang, K. E. Espelie, J. W. Todd, A. K. Culbreath, R. N. Pittman and J. W. Demski. 1993. Cuticular lipids from wild and cultivated peanuts and the relative resistance of these peanut species to fall armyworm and thrips. Jour. of Ag. and Food Chemistry 41:814-818.

Chamberlin, J. R., A. K. Culbreath, J. W. Todd and J. W. Demski. 1993. Detection of tomato spotted wilt virus in tobacco thrips (Thysanoptera: Thripidae) overwintering in harvested peanut fields. J. Econ. Entomol. 86:40-45.

Chamberlin, J.. R. A. K. Culbreath, J. W. Todd and J. W. Demski. 1993. Acquisition of tomato spotted wilt virus by tobacco thrips from asymptomatic and symptomatic peanut. Presented at Southeastern Branch Entomological Society of America 7-10 March at Little Rock, Arkansas.

Culbreath, A. K., J. W. Todd, J. W. Demski and J. R. Chamberlin. 1993. Aggregation of symptomatic and asymptomatic TSWV - infected peanut plants. Presented at Southern Division of American Phytopathology 31 Jan. - 04 Feb. 1993 at Tulsa, Oklahoma.

Li, Z., R.L. Jarret, R.N. Pittman and J.W. Demski. 1994. Organogenesis from cultured seed explants of peanut (*Arachis hypogaea* L.) using thidiazuron. *In vitro* Cellular and Developmental Biology-Plant 30:187-191.

Li, Z., J.W. Demski, R.L. Jarret and R.N. Pittman. 1994. Development of transformation systems for wild and cultivated groundnuts for the integration of virus coat protein genes: p 16-17 in; Working Together on Groundnut Virus Diseases. ICRISAT, Pdatacheru 502324, India.

Todd, J.W., A.K. Culbreath and J.W. Demski. 1994. Insect vectors of groundnut viruses: p.23-24 in; Working Together on Groundnut Virus Diseases. ICRISAT,k Patancheru 502324, India.

Demski, J.W. and C.M. Deom. 1994. The peanut CRSP virus project: p 28 in; Working Together on Groundnut Virus Diseases. ICRISAT, Patancheru 502324, India.

Sreenivasulu, P., D.L. Pinnow and J.W. Demski. 1994. Comparison of five ELISA procedures for detection of peanut stripe virus. Univ. of Georgia Research Report 628, 9 pp.

Olorunju, P.E., S.M. Misari, and J.W. Demski. 1994. Performance of introduced early - and late maturing groundnut varieties at Samaru, Nigeria. IAN 14:16.

Sreenivasulu, P., and J.W. Demski. 1994. Isolation and separation of common peanut potyviruses. Int. J. Trop. Plant Dis. 10:241-251.

Sreenivasulu, P., and J.W. Demski, D. E. Purcifull, R. G. Christie and G. R. Lovell. 1994. A potyvirus causing mosaic disease of sesame (*Sesamam indicum*). Plant Disease 78: 95-99.

Culbreath, A.K. J.W. Todd, W.D. Branch, S.L. Brown, J.W. Demski and J.P. Beasley Jr. 1994. Effect of new peanut cultivar Georgia Browne on epidemics of spotted wilt. Plant Diseases 78:1185-1189.

LI, Z., R.L. Jarret and J.W. Demski. 1995. Plant regeneration from protoplasts of *Arachis* spp. pp. - in Plant Protoplasts and Genetic Engineering. Y.P.S. Bajaj editor, Springer verlag, Berlin, New York. (In press)

Olorunju, P.E., C. W. Kuhn, O.A. Ansa, S.M. Misari, and J.W. Demski. 1995. Mechanical inoculation procedure to screen for resistance to groundnut rosette virus in groundnut. Peanut Science 21: 26-58.

Camann, M.A., A.K. Culbreath, J. Pickering, J.W. Todd and J.W. Demski. 1993. Spatial and temporal patterns of spotted wilt epidemics in peanuts. Phytopathology 85:879-885.

Yang,G., K.E. Espelie, J.W.Todd, A.K. Culbreath, R.N. Pittman and J.W.Demski. 1995. Cuticular lipids from cultivated and wild peanut species and their effects on feeding by fall armyworm, *Spodoptera frugiperda*. Peanut Science 22: 49-54.

LI, Z., R.L. Jarret, M. Cheng, and J.W. Demski. 1995. Improved electroporation buffer enhances transient gene expression in *Ara*chis hypogaea protoplasts. Genome. (In press)

Prasada Rao, R.D.V.J., G. PIÓ Ribeiro, R. Pittman, D.V.R. Reddy, and J.W. Demski. 1995. *In vitro* culture to eliminate peanut stripe virus from peanut seed. Peanut Science 21: 54-56.

Satyanarayana, T., S. Mitchell, S. Brown, S. Kresovich, D.V.R. Reddy, R. Jarret, and J.W. Demski. 1995. The complete nucleotide sequence of the S RNA segment of peanut bud necrosis *Tospovirus* (Bunyaviridae) representing serogroup IV. Journal of General Virology. (In Press)

Cheng, M., J.W. Demski, R.L. Jarret, Z. Li, and R.N. Pittman. 1994. Transformation of peanut (*Arachis hypogaea* L.) using agrobacterium tumefaciens. Agronomy Meeting Nov. 1994. (Abstr.)

Cheng, M., A. Xing, Z. Li, R.L. Farret, and J.W. Demski. 1995. Production of fertile transgenic peanut plants using *Agrobacterium tumefaciens* and the expression and inheritance of foreign genes in transgenic peanut plants. 1995 Congress on In Vitro Biology. Denver, Co, May 20-24, 1995. (Abstr.)

Li, Z., A Xing, M. Cheng, R.L. Jarret, R.N. Pittman, and J.W. Demski. 1995. Asymmetric somatic hybridization via protoplast fusion in peanuts. 1995 Congress on In Vitro Biology. Denver, CO, May 20-24, 1995. (Abstr.)

Li, Z., A Xing, M. Cheng, R.L. Jarret, and J.W. Demski. 1995. Protoplast-mediated transformation of peanut (*Arachis hypogaea*) for virus resistance. 1995 Congress on In Vitor Biology. Denver, CO, May 20-24, 1995. (Abstr.)

TRAINING:

A two week training course on viruses infecting peanut and other legumes was held in Khon Kaen, Thailand during February 1995. This training was supported by the Peanut CRSP, FAO, ICRISAT and a Belgium project. It was held at Khon Kaen University with participant from 10 S.E. Asian countries. Sopone Wongkaew, Thailand and J. W. Demski, USA were the two instructors representing the Peanut CRSP.

PLANS FOR 1995-1996:

Plans are being completed for a training course to be held at the Oil and Botein Seed Center, Potchefstroom, S. Africa from March 25 to April 03, 1996. A participant from 10 African countries will be the trainees.

A survey will be conducted in East Africa to test susceptible and resistant peanut lines for the presence of the groundnut rosette virus, the groundnut rosette assistor virus and the satellite to GRV using antisera or probes that are appropriate. Also individual migrating aphid will be tested for GRV, GRAV and the satellite.

Constraints Associated with Peanut Production, Processing and Use

Environmental

Dependence on chemicals to control insect pests and diseases.

Early infected peanut with groundnut rosette provides a source of virus for other peanuts. When aphids colonize these infected peanuts, the virus is readily carried to surrounding plants inducing epidemics. A control practice to prevent this virus spread is to apply insecticides. The use of groundnut rosette resistant peanut cultivars eliminates the use of insecticides for virus control.

Peanut cultivation in the Sahel region of West Africa may be related to southward expansion of the Sahara Desert.

The peanut virus project is moving groundnut rosette resistant genes into early maturing and drought resistant peanut lines. This will permit the production of peanut in the northern Sahel area.

Inadequate diversity in cultivated peanut germplasm grown in developing countries and the US.

The cultivated peanut is especially lacking genetic diversity for disease and insect resistance. Two approaches are being developed to overcome this problem.

a. Virus genes that code for the production of virus coat protein have been isolated and are being introduced (transformed) into the peanut genomic base. Peanuts having this gene should have some degree of resistance against its homologous virus. This system serves as a model for the insertion of genes controlling many other factors such as anti-aflatoxin, agronomic characteristics, herbicide resistance, etc.

b. We have pioneered in the area of protoplast regeneration and protoplast fusion followed by regeneration. This permits the moving of genetic material from and between peanut species.

Socioeconomic

Low productivity or yield loss from disease, drought, insects and nematodes.

The breeding project in Nigeria is moving the double recessive genes that confer groundnut rosette resistance into better yielding lines as well as lines with earliness and drought tolerance.

In the U.S., the program to transform and regenerate peanut with virus coat protein genes will produce resistant plants from disease loss. It will also provide the mechanism by which other genes can be introduced to retard the yield loss from many other constraints such as insect and nematodes.

Local resource management situations may prevent efficient production and use.

Insufficient local food supply or inadequate food technology prevents growers, processors, or consumers from exploiting the full potential of peanut because it is not considered as a primary food source.

Peanut production in Nigeria for both food and export was once a flourishing agricultural endeavor. Peanut was part of the primary food source. Several factors led to the decline of peanut production in this area of the world; one factor being epidemics of groundnut rosette. In an area where crop production must take place from June to September (when it rains), the growers prefer to grow crops that are not endangered by total loss (rosette epidemics of peanut can cause total loss). Providing rosette resistant peanut seed can overcome this constraint.

Information not available to potential beneficiaries of new peanut production and use technologies.

One objective for the virus project in Nigeria is to have demonstration plots with rosette resistant and susceptible plants to show growers the value of using seed of resistant lines. Dr. Steve Misari, Head of Extension at IAR is a co-principal cooperator.

Training courses of two week duration have been given to participants from many developing countries to update and give training on many aspects of peanut production, use technologies and virus disease control.

Economic losses resulting from mycotoxin contamination.

Health and nutrition

Health hazard from mycotoxins in contaminated peanut.

Methods needed to incorporate peanut and peanut products into safe and nutritious processed foods.

Research capacity and tools

Inadequate number of trained research and support personnel in host countries.

Mandhana Sukorndhaman from Thailand trained and received a PhD at the University of Georgia and returned to Thailand where she works. Phindile Olorunju trained and received a PhD at the University of Georgia. She returned to Nigeria and is the Co-principal investigator on the Peanut CRSP project.

Araceli Pua received 16 months training at the University of Georgia after which she returned to the Philippines to work at the University in Los Banos.

Research tools using molecular biology are not readily applicable to peanut. These tools enable efficient and effective germplasm improvement with respect to resistance to pests and diseases, tolerance to abiotic stresses, seed quality and quantity, and adaptation to growing season. Thus, molecular genetic tools will enhance our capacity to overcome constraints related to environment, economic growth, and human health and nutritions.

The use of molecular biological tools for peanut enhancement have been greatly advanced because of the Peanut CRSP. Some of these advancements are:

a. Regeneration of whole fertile plants from a single protoplast.
b. Model systems to transform peanut with virus coat protein genes.
c. Fusing protoplasts from different peanut species and regenerating the fusion product.
d. Improved electroporation buffer to enhance transient gene expression.
e. *In vitro* inoculation and maintenance of virus cultures.
f. Immunodetection of asymptomatic plants.
g. Organogenesis from cultured seed explants of peanut using thidiazuron.

Table 1. Performance of Early maturing varieties with possible rosette resistance in an advanced groundnut variety trial (Selection trial 1) Samaru, Nigeria 1992-1994.

Serial No.	Entry	Pedigree	Maturity Days	Pod Yield ton/ha	Shelling %	100 Seed mass (g)	Seed[b] Color
1.	ICGS 45		110	1.51[a]	62	36.9	T
2.	ICGV 88709	SPI X RMP 91	110	1.23	69	27.2	T
3.	ICGV 88710	Egret X RMP 91	110	1.55	67	36.1	T
4.	ICGV 88711	Mani Pinta X RMP 91	110	1.79	65	38.7	Vg
5.	48-115B		90	1.11	58	39.2	T
6.	M359-79	RMP12X(4750.70X3520.71	110	1.84	58	39.2	T
7.	176-85A		110	1.64	71	26.9	T
8.	1691-79		110	1.83	57	29.8	T
9.	M669.80		110	1.36	62	36.6	R
10.	M563.78		110	2.14	58	40.0	T
11.	UGA 2	RMP12 X ICGS-56(E)	110	1.73	65	31.3	Vg
12.	UGA 3	RMP12 X 55-437	110	2.04	67	37.8	Vg
13.	UGA 4	RMP12 X M1204.78I	110	2.24	67	35.9	Vg
14.	UGA 5	RMP12 X JL 24	110	1.80	57	38.9	Vg
15.	UGA 7	RMP12 X RRB	110	1.69	73	30.6	Vg
16.	UGA 8	RG 1 X ICGS 56(E)	110	1.49	65	40.1	T
17.	UGA 9	RG 1 X 55-437	110	2.05	54	41.3	T
18.	UGA 10	RG 1 X M1204.78I	110	1.91	67	38.6	T
19.	UGA 11	RG 1 X JL 24	110	1.29	66	30.0	T
20.	UGA 13	RG 1 X RRB	110	1.92	69	32.9	T

[a] Yield averaged over three years.
[b] T = Tan, Vg = Variegated, R = Red

Table 2. Reaction of F_1 segregating populations to infection by groundnut rosette virus at Samaru, Nigeria.

Entry	Genotype	Rosetted Plants	Total Number Plants	Percent Rosetted
1.	ICGM 751/754 X 55-437	25	490	5.1
2.	ICGM 751/754 X ICGV 86055	418	920	45.4
3.	ICGM 751/754 X ICGV 86061	327	1019	36.5
4.	ICGM 751/754 X ICGV 86105	11	260	4.2
5.	ICGM 751/754 X ICGV 87922	179	994	18.0
6.	ICGM 751/754 X Chico	127	327	38.8
17.	UGA 3 Resistant Control	0	229	0.0
18.	48-115 B Susceptible Control	85	85	100.0

Table 3. Effect of potassium glutamate (KGlu) concentration on protoplast viability and GUS expression after electroporation.

Kglu (mM)	Ionic conductivity (µmho)	Time constant (msec)	Protoplast viability (%)	GUS activity (pmol MU/ min per 10^6 proptoplasts)
10	1710	119	2.2	16.5
30	2470	85	6.7	1033.3
50	3250	68	11.1	3847.9
70	3965	59	36.7	4009.3
100	5150	48	50.0	3308.6
150	6815	35	75.5	705.7
250	9940	27	71.1	451.1

Note: Fifty micrograms of DNA was used in each treatment. Protoplast viability rate and GUS activity values represent the mean of three sample observations per treatment from two experiments.

Table 4. Regeneration of shoots from protoplast-derived callus colonies of *A. paraguariensis* on MS medium supplemented with different concentrations of NAA and BA.

NAA (mgl^{-1})	BA (mgl^{-1})	Mean no. Colonies (total)[1]	Responsive colonies	Shoots (total)[2]	Shotts/ colony
0	0.0	15	0	0	0.0
0	0.5	15	13	79	6.1
0	1.0	15	13	54	4.2
0	3.0	15	12	59	4.9
0.1	0.5	15	15	85	5.7
0.1	1.0	15	15	139	9.3
0.1	3.0	15	9	90	10.0
0.5	0.5	15	10	52	5.2
0.5	1.0	15	15	134	8.9
0.5	3.0	15	13	109	8.4

[1] Five protoplast-derived colonies were inoculated in one Petri plate. Each treatment contains 3 replicate plates.

[2] Data were collected 50 d after inoculation.

Table 5. Inheritance of GUS activity in progeny of T_0 transgenic peanut plants.

T_0 plant	No. of T_1 plants tested	GUS+	GUS-	3:1 Segregation‡ X^2	P
1-4	14	12	2	0.38	0.54
1-5	8	7	1	0.90	0.35
1-9	18	14	4	0.07	0.79
1-10	12	7	5	1.77	0.18
1-13	20	3	17	38.40	0.00
1-44	10	10	0	3.33	0.07
1-45	13	0	13	39.00	0.00
17-1	24	19	5	0.22	0.64
12-1	28	20	8	0.19	0.66
12-2	8	6	2	0.00	1.00

‡ Goodness of fit to 3:1 ratio. df=1

Annual Report
July 1994 - June 1995

AAMU/FT/BF

An Interdisciplinary Approach to OptimumFood Utility of the Peanut in SAT Africa

Alabama A&M University
University of Ouagadougou, Burkina Faso
Food Research Institute, Ghana

Principal Investigators: M. Elena Castell-Perez (Alabama),
Alfred S. Traore (Burkina Faso) and Kafui Kpodo (Ghana)

INTRODUCTION

Work on improved utilization of peanuts for human consumption in Semi Arid Tropical Africa (Burkina Faso and Ghana) continued. Efforts consisted on (a) increasing utilization of peanuts into more refined/processed forms (USA and Burkina Faso); (b) improving packaging of peanuts and peanut products to increase shelf-life (Burkina Faso); (c) utilizing peanut flour (after extraction of oil) to increase protein value of cereal-based foods (USA and Burkina Faso); (d) improving postharvest handling methods to reduce aflatoxin problem (Burkina Faso); and (e) identifying level of aflatoxin contamination at farm and processing level (Ghana).

Major Accomplishments

1. Weaning foods: (USA) The use of peanut paste in infant weaning foods was evaluated in terms of physical characteristics. Addition of this ingredient to mixes of cowpea, pearl millet, and teff flours decreased the water absorption index. Roasting of peanuts at 160°C for 40 minutes decreased the WAI by 25%. Research on roasting procedures was conducted to determine optimum conditions in terms of color and aroma. Qualitative and quantitative nutritional analyses of the composite flours are in progress. Good formulas of weaning foods using maize, sorghum and rice, peanut flour and milk have been developed in Burkina Faso. The product has improved nutritional value (18-20% protein, 10-12% fat, 2-3% minerals) and higher energy density.

2. Development of reduced-fat peanut butter: Optimum conditions (roasting, oil extraction procedures, type of fat replacers) were established for manufacture of a peanut paste with 50% less fat than commercially available products in the USA. Sensory characteristics (color, texture and taste), viscosity and storage stability (shelf-life) of product were determined. Work is in progress to evaluate the flavor quality of the paste using Gas Chromatography techniques.

3. Extruded snacks: The nutritional value, physical characteristics and shelf-life of an extruded peanut-sorghum wafer were determined. Sensory work is in progress.

4. Effect of fortification on dough quality of cereal-based foods (USA): Studies on proximate composition and rheological properties of peanut-fortified corn flour and dough indicate a considerable improvement on dough spreadability and shelf-life with supplementation up to 30% with defatted peanut flour.

5. Peanut paste quality and packaging (Burkina Faso): Nutritional and microbiological quality of peanut paste from several regions of Burkina Faso was analyzed. Quality of traditionally made paste was generally inferior to the product manufactured by a local industry. results encourage the development of industrial processing plants for manufacture of peanut products. The optimum package size and material were identified by scientists at the Universite de Ouagadougou and information delivered to the peanut paste factory CITEC-HUILERIE.

6. Fortification of traditional or staple foods (Burkina Faso): Scientists at the University of Ouagadougou completed research on processing conditions for peanut-fortified staple foods such as "toe" and "basi".

7. Studies on aflatoxin contamination of peanuts: Procedures for isolation and preparation of components for aflatoxin growth inhibition of a strain of Aspergillus flavus with an extract of Allium sativum are in progress.

8. Identification of problems with postharvest handling of peanuts in Ghana: A survey questionnaire was improved and utilized in all regions of Ghana. Data on aflatoxin contamination and postharvest practices were collected.

Expected impacts of the project

The major constraint in increasing peanut utilization in SAT Africa is the unavailability of peanut in processed forms: packaged roasted peanuts, peanut butter, defatted peanut flour, peanut milk, candies and other confectioneries at affordable costs. Secondly, contamination of peanuts with molds, insects, and mycotoxins further limits the utility for human consumption.

Implementation of research results have lead to better utilization of peanuts in Burkina Faso (weaning foods and staple foods, reduced aflatoxin contamination). Specifically, results have lead to improvement of methods of storage and packaging, processing and development of a variety of products in the country. It is expected that this knowledge will be transferred to other SAT African countries via training and/or workshops through the scientists at the Universite de Ouagadougou because the Peanut CRSP project has certainly enhanced the capability in research at the University of Ouagadougou to address these constraints.

Results from research conducted on the use of Allium sativum as an inhibitor of aflatoxin production on peanuts during storage will have a tremendous impact on the health conditions of the local population. The same objectives will be applied to Ghana where the conditions under which peanuts and their products are sold in the markets predispose them to rancidity development and its associated nutritional and acceptability problems as well as microbiological and mycotoxicological implications. This emphasizes the urgent need to review and upgrade the existing processing methods and quality of traditional peanut products in the country.

Goals

General Goal

To foster interdisciplinary (nutrition, food science, social and economic) institution-based linkages between U.S. and LDC scientists serving major peanut producing and consuming populations of the Semi-Arid Tropic regions of Africa for the purpose of optimizing the food utility of the peanut.

Specific Goals

Specific goals of the project are consistent with the general goal of the peanut CRSP to develop collaborative research and development programs on the peanut between scientists at Alabama A&M University, the University of Ouagadougou in Burkina Faso, and the Food Research Institute in Accra, Ghana.

ORGANIZATION

USA (Alabama A&M University)
Dr. M. Elena Castell-Perez, Principal Investigator, Department of Food Science, Normal, AL, Engineer.
Dr. John C. Anderson, Co-Principal Investigator, Department of Food Science, Normal, AL, Engineer.
Mr. Simon Ogutu, Research Technician, Department of Food Science, Normal, AL, Food Scientist.

Burkina Faso
Dr. Alfred S. Traore, Principal Investigator, Food Biochemist.
Mr. Solibo Some, Cooperator, Entomologist.
Dr. Philippe Nikiema, Cooperator, Food Technologist.
Dr. Albert Ouedraogo, Cooperator, Entomologist.
Mrs. Lactitia Ouedraogo, Cooperator, Food Technologist.

Mr. Alain Traore, Research Assistant, Food Scientist.
Mr. Francois T. Ouedraogo, Cooperator, Food Technologist.
Mr. Philippe Sankara, Cooperator, Phytopathologist.
Mr. Darouda G. Traore, Research Technician, Food Scientist.

Ghana
Mrs. Kafui Kpodo, Principal Investigator, Food Scientist.
Dr. Wisdom A. Plahar, Co-PI, Food Scientist.
Ms. Hodare-Okae, Cooperator, Food Microbiologist.
Ms. N.T. Annan, Cooperator, Nutritionist/Biochemist.
Mr. R.K. Adjei, Cooperator, Agricultural Economist.
Mr. C.K. Gyato, Cooperator, Agricultural Engineer.

Graduate students and research projects for thesis

USA
1. Mr. Surendra K. Singh - Thesis Topic: Flavor and Rheology of Reduced-Fat peanut Paste. M.S. Degree expected December 1995.
2. Ms. Linda Griffith - Dissertation Topic: Precooked weaning foods based on cereal/peanut/legumes composites. Ph.D. expected December 1995.
3. Ms. Nelly Duarte - Dissertation Topic: Development of peanut fortified non-extruded snack foods. Ph.D. expected December 1995.
4. Mr. Rolfe Bryant - Dissertation Topic: Control of oligosaccharide production in peanut. Ph.D. expected December 1995.

Burkina Faso
5. Mr. Simeon Nanema - Thesis Topic: Improvement of nutritional quality of toe on fortification with peanut proteins. Doctorat Degree expected December 1995.

Accomplishments and approach in detail

Comparative Study of Nutritional and Microbiological Quality of Peanut Pastes from Burkina Faso:

Nutrient composition, aflatoxin levels and microbiological contamination of peanut paste manufactured in Burkina Faso were compared. Pastes were either manufactured by the traditional method of using stone slabs or a small plate mill and then stored in containers or industrially made pastes (manufactured by CITEC-HUILERIE). The traditional pastes were collected from markets in several cities of Burkina Faso. Results indicate that proximate composition of traditional pastes varied slightly from one city to the other. Protein content (20.49-23.32%) of traditional pastes was significantly lower ($P<0.005$) than industrial pastes (30.26%). Industrial pastes were also less susceptible to rancidity (given by peroxide values 1.5-3.9 times lower than for the traditional pastes). Aflatoxin levels in traditional pastes (25.38-31.36 ppb) were considerably higher than in industrial pastes (19.13 ppb). Microbiological studies revealed noticeable contamination of traditional pastes. Results suggest that better storage, processing and handling procedures should be adopted by local manufacturers to increase product quality and shelf-life. The need for better packaging is also needed to guarantee product quality, extended shelf-life and therefore better promotion and commercialization of the paste.

MATERIALS AND METHODS

Sampling

Peanut paste samples were collected at five (5) markets from four (4) cities located in different geographical zones of peanut production: Banfora (South and Southwest), Bobo-Dioulasso (West and Northwest), Ouagadougou (Center), and Tenkodogo (East and Northeast). In each market, samples (500g) were collected from three (3) sellers in January, March and June. Samples were placed into plastic bags and taken to the laboratory for analysis. A total of 180 samples was evaluated. Market samples were manufactured using the traditional

method of peeling, roasting and milling the peanut kernels using two stones. Paste is then placed into big metal containers without further protection from the environment.

Samples from CITEC-HUILERIE were manufactured by following a series of steps such as cleaning, sorting, blanching, roasting and grinding.

Chemical Analyses

Moisture, free fatty acids, crude fiber, carbohydrate, fat, protein and ash content were determined using standard AOAC methods. Energy density was calculated using FAO conversions for fat, protein and carbohydrate. Aflatoxin B_1 was monitored using the inoculum method. Vitamin C was determined with the 2,6-dichloroindophenol titrimetric method.

Microbiological Control

Total microflora of samples were numbered using petri plates technique with standard agar methods. Coliforms and clostridia were determined on specific media. Staphylococci, yeast and mold counts were performed using standard procedures.

RESULTS

The proximate analysis of the collected peanut paste samples is presented in Table 1. Results show that moisture content in pastes from Bobo-Dioulasso and Ouagadougou was 23-25% higher than the other pastes. This marked difference can be attributed to the fact that people add water to their product to increase the quantity (volume). Protein content of the industrial paste was considerably higher (22.9-32.3%) and carbohydrate content was lower (46.5-64.5%) than for the traditional pastes. This confirms the common use of product adulteration practices by adding sorghum or cereal flour to the product to increase product quantity. The rancidity of pastes as indicated by the peroxide value indicate that traditional pastes became rancid more rapidly. This was expected due to the improper storage and package of the paste. Aflatoxin content of the various traditional unpacked pastes were comparable (25.38-31.36 ppb) and much higher than for the industrial paste (19.13 ppb).

Microbiological quality of traditional pastes was generally poor (Table 2). In contrast, the level of microbial contamination in the industrial paste was negligible. These results point out the high health risk at which consumers are exposed by purchasing these products.

In summary, the research showed that industrial processing of peanut paste yields a product with good nutritional quality and free of contamination by microbes and/or aflatoxins. Therefore, local traditional peanut paste manufacturers must be educated on practices to improve their products.

Table 1. Proximate Composition of Peanut Pastes Sampled in Different Cities of Burkina Faso

Parameter	Mean values of parameters on peanut pastes at various locations[1]				
	B.[a]	B.-D.[b]	Ouaga[c]	Tenk.[d]	CITEC-HUILERIE[e]
Moisture (%)	0.77	1.08	1.06	0.86	0.80
Free fatty acids (%)	0.60	0.69	0.54	0.30	0.57
Fat (%)	51.26	49.30	49.15	50.29	51.60
Protein (%)	20.63	23.32	22.04	21.48	30.26
Carb. (%)	25.06	22.31	24.48	23.26	15.23
Fiber (%)	1.80	1.44	2.33	2.50	2.20
Ash (%)	2.59	2.46	2.47	2.47	2.55
Heat content (kcal/g)	6.33	5.85	6.02	5.66	6.11
Vitamin C (mg/100g)	14.00	14.80	13.07	13.86	13.71
Peroxide value (mMO_2/kgoil)	11.93	16.75	35.80	18.03	9.08
Aflatoxin B_1 (ppb)	31.36	30.31	26.33	25.33	19.13

[1] Mean values from 45 samples
[a] Banfora, [b] Bobo-Dioulasso, [c] Ouagadougou, [d] Tenkodogo
[e] Mean values from 15 samples

Table 2. Microbial Contamination in Peanut Paste Samples from Different Cities of Burkina Faso

Microorganism	Mean values of microbial counts on peanut pastes at various locations[1]				
	B.[a]	B.-D.[b]	Ouaga[c]	Tenk.[d]	CITEC-HUILERIE[e]
Total micro-flora ($.10^3$/g)	1529.0	1130.0	1239.0	1896.0	0.00
Coliforms ($.10^3$/g)	1.1	48.9	2.2	21.1	0.00
Staphylococci ($.10^3$/g)	114.5	82.9	672.6	616.6	0.00
Salmonella/ Shigela ($.10^3$/g)	0.7	2.6	0.6	16.1	0.00
Clostridia ($.10^3$/g)	53.7	169.9	280.2	128.2	0.00
Yeast/mold ($.10^3$/g)	268.5	79.5	12.1	80.5	0.00

[1] Mean values from 45 samples
[a] Banfora, [b] Bobo-Dioulasso, [c] Ouagadougou, [d] Tenkodogo
[e] Mean values from 15 samples

Dough Quality:

Storage of dough presents a big challenge in the food industry because the stability of frozen dough during storage, and during freezing-thawing remains a problem. The purpose of this research was to analyze the effect of peanut supplementation on corn (masa) flour storage quality. Studies on rheological changes of dough due to temperature and composition are currently in progress.

MATERIALS AND METHODS

Dry masa flour (DMF) was fortified with chickpea flour (CPF) (up to 30%) and partially defatted peanut flour (PDPF) (up to 25%). Six combinations of composite flours were selected based on good dough manageability properties for processing. This was determined by handling and sheeting the fresh dough, the frozen-thawed dough and by making a tortilla-like product. The masa and composite dough were stored at -18°C for 6 months and defrosted for analysis. Water activity of the dough during storage was monitored using a Rotronic measuring system. textural characteristics (compression force) were determined using an Instron Universal testing Machine for fresh and stored samples (every two weeks for up to 6 months). Proximate analysis of samples was done using standard procedures.

RESULTS

Fortification of dry masa flour improved the textural characteristics of the dough during frozen storage (lower values of compression force which indicate a softer product versus the hard texture of masa dough). Dough manageability (cohesiveness) also improved with fortification. Masa dough lost plasticity during storage due to

greater water loss as compared to the composite dough which remained acceptable for processing. Proximate analysis of flours showed a desirable increase in protein content (8 to 16%). However, fortification also increased fat (3-8%) and moisture content (8-14%, w.b.). Results indicate that fortification of dry masa flour produced a dough with acceptable quality (textural) after 6 months of frozen storage.

Field Survey on Peanut Production in Ghana:

The objective of the survey was to study peanut production in the major peanut growing areas of Ghana. The survey covered the Ashanti, Brong-Ahafo, Northern, Upper-East, and Upper-West regions. Information obtained from the survey included the general background of peanut farmers, practices for land preparation, planting, harvesting, threshing (stripping), drying, shelling, storage, and general problems which hinder peanut production in Ghana. A summary of results from fifty farmers is given below:

Peanut is grown by small-scale farmers with an average planted area of 3.5 acres. Two thirds of the farmers had not had any formal education. Severeal varieties are planted with the most widely cultivated ones preferred because they are easy to harvest (by manual pulling from the soil), early maturing and high yielding. Except for land preparation which was carried out by both manual means and in some cases by use of tractors, all other operations such as planting, harvesting, threshing, and sorting were manually done.

Drying of peanuts after harvesting was carried out directly in the sun either on the farm or at home. Most farmers store peanuts unshelled in jute sacks or in traditional storage structures until they are ready to sell. Storage could be for as long as 9 months. Only 8% of farmers interviewed used shellers. 35% of farmers did not sort peanuts and those who sorted did so to get a good market price for their produce. All the farmers interviewed were completely ignorant about the aflatoxin problem in peanuts.

Buyers of peanuts at the farmgate are local traders and middlemen from the urban centers. Major problems mentioned by peanut farmers were unreliable rainfall, poor soil fertility, and lack of farming inputs for land preparation, harvesting, threshing, shelling, and high labor cost. All the farmers interviewed were willing to adopt new equipment for peanut production if offered to them on a hire-purchases basis.

Studies on Existing Traditional Processing Procedures for Peanut Butter in Ghana:

A nationwide survey covering the ten regions of Ghana was conducted to study the existing traditional methods for the preparation of peanut butter. The major unit operations identified were roasting, dehulling, winnowing, milling, and packaging. Analysis of data and samples collected is in progress.

Quality Evaluation of Traditional Peanut Butter Samples in Ghana:

Twenty peanut butter samples from the Eastern and Central regions of Ghana were analyzed for aflatoxin contamination (B_1, B_2, G_1 and G_2) using High performance Liquid Chromatography (HPLC). Generally, extremely high levels of aflatoxin were detected in the samples. Only two out of the twenty samples were aflatoxin-free (detection limits for aflatoxin B_1 and B_2 of 0.04 μg/kg and for G_1 and G_2 of 0.06 μg/kg). All the positive samples had levels above the WHO/FAO/UNICEF maximum permissible limit of 30 μg/kg for protein supplements. Total aflatoxin levels for positive samples from the Eastern region (Table 3) ranged from 47.7 to 2024.1 μg/kg. Total aflatoxin level for the Central region (Table 4) was 78.0 to 2013.9 μg/kg. For the Eastern region generally all four types of aflatoxin were detected suggesting contamination by both *Aspergillus flavus* and *Aspergillus parasiticus*. In the case of the Central region, however, aflatoxin G_1 and G_2 were less frequently detected suggesting a dominance of *Aspergillus flavus* (responsible for the production of aflatoxin B_1 and B_2) over *Aspergillus parasiticus*. Analysis of samples from the remaining regions is in progress.

Ten samples of peanut butter from the Eastern region were analyzed for moisture, protein, fat, and free fatty acids. Moisture levels ranged from 0.7% to 3.0%. This may be due to differences in the extent of roasting. Protein and fat contents were characteristically high ranging from 27% to 36% and 40% to 53%, respectively. All but one

peanut butter samples had free fatty acid (FFA) values within the range of 0.5% to 1.3% (as oleic). One sample had a FFA content of 5.8% which was extremely high. Most oils produce noticeable rancidity to the palate when the FFA is about 0.5-1.5%.

The microbiological quality of twenty five samples from the Eastern, Central and Western regions was evaluated. Microorganisms isolated included *Bacillus spp*, *Candida sp*, *Aspergillus niger*, *A. ochraceus*, *A. candidus*, *A. flavus*, and *A. parasiticus*. Other molds isolated in smaller numbers were *Penicillium spp*, *Torula sp.*, and *Monilia sp.*. However, *Aspergillus spp.* were found to be the most prevalent and accounted for about 80% of the total mold population of the samples. Bacterial count of the samples ranged from 120 to 9.3×10^5 CFU while mold count ranged from <10 CFU to 5.6×10^5 CFU. No pathogenic bacterial organisms (*E. coli*, *Staphylococcus aureus* and *Salmonella spp.*) were isolated.

Table 3. Aflatoxin Content of Peanut Butter Samples From the Eastern Region of Ghana

Sample Code	Aflatoxin Level (µg/kg)				
	B_1	B_2	G_1	G_2	Total
Nkawkaw (ER)					
EN-1	53.0	40.5	15.6	ND.	109.1
EN-2	56.4	18.0	ND.	ND.	74.4
EN-3	291.8	115.7	ND.	ND.	407.5
EN-4	341.4	315.5	273.9	212.3	1143.1
EN-5	ND.	ND.	ND.	ND.	ND.
Koforidua (ER)					
EK-1	582.6	177.1	434.1	119.3	1313.1
EK-2	1054.2	421.0	274.8	274.1	2024.1
EK-3	286.7	158.6	368.3	195.2	1008.8
EK-4	29.1	18.6	ND.	ND.	47.7
EK-5	220.4	132.8	82.3	67.7	503.2

ND = None detected

Table 4. Aflatoxin Content of Peanut Butter Samples From the Central Region of Ghana

Sample Code	Aflatoxin Level (µg/kg)				
	B_1	B_2	G_1	G_2	Total
Cape Coast (CC)					
CC-1	1336.7	494.2	123.0	60.0	2013.9
CC-2	178.4	53.8	ND.	ND.	232.2
CC-3	639.0	197.7	ND.	ND.	606.6
CC-4	562.7	73.3	ND.	ND.	636.0
CC-5	140.2	59.9	ND.	4.2	204.3
Mankesim (CM)					
CM-1	261.7	92.0	ND.	ND.	353.7
CM-2	65.5	12.5	ND.	ND.	78.0
CM-3	639.0	197.7	ND.	93.6	930.3
Swedru (CS)					
CS-1	194.4	51.6	ND.	ND.	251.0
CS-2	ND.	ND.	ND.	ND.	ND.

ND = None detected

Training Accomplishments:

Degree

1. Mr. Bin Pan - Thesis Topic: Effect of peanut flour supplementation on textural and rheological changes of wheat dough during microwave baking. M.S. Degree, July 1994.

2. Mr. Iwan Surjawan - Thesis Topic: Physical and functional properties of a sorghum-peanut extruded wafer product. M.S. Degree, December 1994.

3. Ms. Wichayada Intaratip - Thesis Topic: Nutritional assessment of sorghum and peanut-based wafers for military rations. M.S. Degree, May 1995.

4. Mr. Brent Jones - Thesis Topic: Storage stability of sorghum-peanut wafers. M.S. Degree, June 1995.

5. Mr. Rufin Simde (Burkina Faso) - Thesis Topic: Improvement of shelf-life of peanut products (roasted peanut kernels and locally made peanut pastes). Doctorat Degree (December 1994).

Publications and Presentations:

A. Manuscripts:

1. Castell-Perez, M.E. and Mishra, A. 1995. Flow behavior of regular and peanut-fortified idli batters. J. of Texture Studies 26(3): 273-279.

2. Pan, B. and Castell-Perez, M.E. 1995. Texture and viscoelasticity of canned biscuit dough during microwave and conventional baking. Submitted to J. Food Processing Engineering.

3. Nikiema, P.A. and Traore, A.S. 1995. Etude de la contamination des graines d'arachide par les aflatoxines produites au cours du stockage. J. Africain de Recherces Chimiques 1(1): 2-14.

B. Presentations:

1. Duarte, N. and Castell-Perez, M.E. 1995. Changes in texture of masa dough due to storage and composition. Institute of Food Technologists, Anaheim, June 3-7.

2. Pan, B. and Castell-Perez, M.E. 1995. Effect of peanut supplementation on texture of wheat dough for microwave baking. Institute of Food Technologists, Anaheim, June 3-7.

3. Castell-Perez, M.E. and Okezie, O. 1995. Aspects of Peanut Processing, Packaging and Utilization in the Semi Arid Tropics of Africa: The Case of Burkina Faso. Peanut CRSP Workshop, Arlington, Virginia, March 29-31.

4. Kpodo, K.A. 1995. Status of aflatoxin research into peanut in Ghana. Presented at the Peanut CRSP/CIRAD/ICRISAT Aflatoxin Collaborative Research Planning Meeting, Accra, Ghana, May.

International Travel:

1. April 1995. Dr. Alfred Traore, principal Investigator from Burkina Faso, and Ms. Kafui Kpodo, Principal Investigator from Ghana, visited Alabama A&M University to discuss research protocols and to revise project objectives. They previously attended the Peanut CRSP Workshop in Arlington, VA, March 29-31.

Research Plan for 1995-1996

Research Plan for 1995-1996

1. Research thrusts/endeavors planned:

U.S.

A. Improvement or modification of peanut processing.

B. Assurance of safety of products considering naturally occurring toxicants and added ingredients with particular reference to mycotoxins in raw and finished products.

C. Improvement of postharvest handling and storage techniques to increase quality of peanuts and peanut products.

BURKINA FASO

A. Studies on peanut products improvement will continue as part of the Food Technology research program at the Universite de Ouagadougou. Priority will be given to packaging issues.

B. Finalize work on use of extracts of *Allium sativum* to suppress growth of *Aspergillus flavus*.

(1) Standardize methods for preparation of crude, distillate and lyophilized extracts of *A. sativum*.

(2) Isolate and identify compounds responsible for growth inhibition of *Aspergillus spp* (done with collaboration from Alabama A&M University's scientists).

(3) Large scale tests of use and effectiveness of extract on peanut seeds during storage. Proper amount of extract and method of application will also be established.

C. Finalize studies on decrease on quality losses due to aflatoxin contamination during storage of peanuts.

(1) Determine effectiveness of locally made plastic dryers to be used on-farm.

(2) Evaluate improvements due to added ventilation to the aforementioned dryers.

GHANA

A. Continue studies on improvement of peanut butter quality and processing methods with assurance of safety considering mycotoxins.

B. Continue work on detoxification strategies and use of plant extracts for inhibition of aflatoxin on peanuts.

(1) Continue studies on application and implementation of plant extracts to test their effect on aflatoxin prevention. Conduct storage tests at different stages of postharvest: fresh peanuts, storage, etc. Monitor changes through time at different conditions.

(2) Explore possibilities of collaborating with scientist at the University of Kumasi.

C. Identify critical problems in production and processing.

(1) Initiate studies on sorting procedures and packaging systems to improve nutritional and organoleptic quality of products as well as to prevent aflatoxin contamination.

(2) Continue studies on development of extruded weaning foods with peanut flour as a component.

2. Training:

A. One graduate student (M.S.) will complete training on characterization of extruded composite flours at Alabama A&M University. Another student will complete his Doctorat in Burkina Faso.

B. Technicians at the Food Research Institute (Ghana).

3. Manuscripts, presentations, releases planned:

A. Manuscripts:

1. "Rheological and Flavor Characteristics of Reduced-Fat Peanut Paste" - to be submitted to the J. texture Studies.

2. "Effect of Processing on Nutritional Quality of Peanut- Based Weaning Foods" - to be submitted to the J. of Nutrition.

3. "Quality Assessment of Peanut-Fortified "Toe" (with Alfred Traore, Burkina Faso) - to be submitted to J. Food Science.

B. Presentations: at the Institute of Food Technologists (IFT) Meeting, American Peanut Research Society (APRES) Meeting and American Society of Agricultural Engineers (ASAE) Meeting.

4. International Travel:

A. U.S. Principal Investigator to visit Ghana and Burkina Faso to review research progress. Two weeks - September 1996.

B. Ghana Principal Investigator to attend the IFT meeting in July 1996.

5. Program Redirections Planned:

(1) Enhanced effort in aflatoxin prevention and postharvest technology problems.

6. Timetable:

All research will continue throughout 1995-1996.

April 1995 - October 1995: Finalize phase out procedures in Burkina faso.

April 1995-April 1996: Determine further approaches for continuing research in Ghana. Provide the Food research Institute with needed equipment and training.

Continue work on improved peanut utilization for development of new and better products at Alabama A&M University.

Progress Toward Resolving Constraints

1. Socioeconomic constraints:

- Insufficient local food supply or inadequate food technology prevents growers, processors, or consumers from exploiting the full potential of peanut because it is not considered as a primary food source.

- Information not available to potential beneficiaries of new peanut production and use technologies.

Through the Peanut CRSP Food Technology project information on technology, packaging and development of new peanut-based products has been presented to local peanut processors in Burkina Faso. Packaging of peanut-based products and consumption of peanut-fortified weaning foods is currently a common practice in that country. More people are directly dealing with peanut products which creates an increase in the activity of the informal sector.

2. Health and nutrition constraints:

- Health hazard from mycotoxin in contaminated peanut.

- Methods needed to incorporate peanut and peanut products into safe and nutritious processed foods.

People have been informed of the importance of hygienic practices during handling and storage of peanuts and peanut products. Alternative methods for prevention of mycotoxin contamination of peanut-based products have been developed and should be put into practice soon. A direct relationship between mycotoxins and liver cancer was established and proper organisms were informed. The nutritional value of staple foods was enhanced by incorporation of peanut flour. Some products are currently in preparation for marketing studies. A survey to identify the levels of contamination in Accra, Ghana, has helped local scientists to establish the application of methodologies to decrease or eliminate the contamination problem.

3. Research capacity and tools:

- Inadequate number of trained research and support personnel in host countries.

The project has furnished equipment to the Food Science Laboratory at the University of Ouagadougou in Burkina Faso and the Food Research Institute in Ghana. Several students have been funded by the project and some are actively working with peanut research problems in Burkina Faso. Field technicians have been hired in Ghana for survey work.

GA/FT/TP

Appropriate Technology for Storage/utilization of Peanut

UNIVERSITY OF GEORGIA - THAILAND AND PHILIPPINES

Larry R. Beuchat, Principal Investigator, UGA
Penkwan Chompreeda, Principal Investigator, Thailand
Virgilio V. Garcia, Principal Investigator, Philippines

INTRODUCTION

Emphasis of the project in FY95 was on developing and transferring processes to utilize peanuts and peanut products in traditional and new food products which would be acceptable in Thailand, the Philippines, other Southeast Asian countries and the U.S. The choice of such products is based in large part on consumer survey response but also on intuition and insight to consumer behavior. Fundamental and applied research was conducted in laboratories of all three collaborating institutions. Utilization of peanut and peanut products in a wide variety of food products has resulted in the development of several items which we believe are highly acceptable and have excellent potential for marketing. Additional marketing studies are needed to ascertain the potential for sales of such products in the Southeast Asian region.

Technology transfer to village scale peanut processors, emphasizing housewives in the Kud-Jub district of Udorn-Thanee Province of Thailand was established in FY95. Cooperation with the Thai Department of Agriculture Extension, Ministry of Agriculture, facilitated this effort.

In the regional and international training area, a course on "International Training: Quality Evaluation and Utilization of Food Legumes" was organized at Kasetsart University, Bangkok, in cooperation with the Department of Technical and Economic Cooperation, Office of Prime Minister under Thai-AID projects. Twenty participants from thirteen countries (Tanzania, Fiji Island, Srilanka, Republic of Maldives, Bhutan, Bangladesh, Sierra Leone, Loas P.D.R., Cambodia, Korea, Vietnam, Nepal and Thailand) attended a 5-week course. Graduate Students are pursuing advanced degrees in food science and technology at Kasetsart University, the University of the Philippines at Los Banos and the University of Georgia while being supported by project funds.

MAJOR ACCOMPLISHMENTS

A. Kasetsart University (KU)(Host Country)

Product concept of a breakfast bar was obtained by a consumer survey. The breakfast bar was formulated to meet 10% of the adult Thai RDA. Product formulations were optimized and sensory evaluation was employed to verify product concepts.

Texturized peanut protein (TPP) was prepared and incorporated into a snack. Quality characteristics of the TPP snack were evaluated. Results of sensory evaluation indicated moderate acceptability of the product.

Tempeh starters were prepared using several substrate formulations. Selection of starter was based on efficiency in producing good quality peanut tempeh. The starter was prepared in larger scale and tested before shipping to housewives at Kud Jub District, Udorn-thanee Province.

Product concept of a mosquito repelling stick based on peanut hulls (shells) was obtained by a consumer survey. Product testing was done and the formula was optimized.

Design criteria for modification of an existing sheller was developed for Khon Kaen 60-1 cultivar. Clearance between the rotating rods of the shelling concave should be 0.5 to 1.0 mm more than the width of the kernel, the clearance between the rubber tire and the concave should be 6.5 to 9.5 mm larger than the width of the kernel, shelling speed should be less than 8.5 m/s and 4.0 m/s for peanuts with moisture content greater than 10% and less than 10%, respectively, and feed rate should be less than 400 kg/hr.

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B. University of The Philippines at Los Banos (UPLB)

The suitability of peanut as an extender of ground pork and beef in meatloaf was evaluated. Results indicate that a formula containing up to 83% defatted peanut or peanut residue has favorable sensory quality.

A peanut nugget snack food and a peanut based cracker were successfully formulated. Crackers containing 70-80% peanut flour were highly acceptable.

The effectiveness of a mold metabolite in controlling the growth of aflatoxigenic aspergilli was determined. Application of the metobolite in the form of a powder appears to enhance the storage stability of refrigerated peanut.

C. Visayas State College of Agriculture (ViSCA)

Supplementation and optimization of Lady Finger formulas with peanut done. Appropriate blends of peanut, wheat flour and cassava flour to give highest levels of sensory quality were defined.

Packaging technologies for baked root crop products extended with peanut were evaluated. Shelf life predictions for various conditions of packaging and storage were established.

Interdisciplinary and integrated efforts to transfer peanut processing technologies to rural women cooperatives were continued. Training, processing management and marketing aspects were given increased and expanded. Successful programs are now clearly in place in two locations with good prospects of expansion to others.

D. University of Georgia

A study was conducted to optimize a honey-peanut butter product (HPNTB). A 3x3 full factorial design representing 3 levels of dry honey (6, 13 and 20% by weight of ground roasted peanuts) and 3 levels of salt (1, 1.5 and 2.0%) was used in the investigation. Ninety-three consumers evaluated 9 samples containing varying proportions of honey and salt and a control peanut butter containing no honey. Ratings were obtained for overall acceptability, spreadability, color, sweetness, saltiness, flavor and texture using a nine-point hedonic scale. The control peanut butter had a texture rating higher than all HPNTB treatments. The treatment containing 6% dry honey and 1% salt did not differ from the control except for texture. Prediction models for spreadability, sweetness, saltiness, flavor, texture and overall acceptability were developed. From prediction equations, a HPNTB formulation containing 7% dry honey and 1% salt would not be significantly different from the control for all attributes except texture.

Ground, blanched, raw and roasted peanuts with varying oil contents were analyzed to demonstrate enzymatic oxidation and autoxidation, respectively. Peroxide value for fresh raw peanuts decreased significantly with decrease in oil content indicating greater stability with lower oil content. Peroxide values of rancid roasted peanut samples likewise exhibited a significant decrease with decrease in oil content. The significantly increasing Oxidative Stability Index time at 110°C for raw and roasted ground peanuts with decreasing oil content supports the assumption of greater chance of autoxidation at higher oil contents. Roasted peanutty and burnt odor, and roasted peanutty, rancid/oxidized, and burnt flavor scores of rancid roasted peanuts significantly decreased with decreasing oil content.

Studies were conducted to determine suitable kinetic models for predicting lipid oxidation in dry roasted peanuts. Models can be used to develop appropriate storage, handling, packaging or pretreatment procedures for maintaining quality of peanuts. Three models were studied, (Maloney, Labuza and Koelsch). Hexanal was a good indicator among various chemical indicators for lipid oxidation. The models by Maloney and Koelsch were most suitable.

Formulation of edible films using peanuts as the main ingredient was studied. The method of forming the film, i.e., forming in on the surface of a heated solution or casting it on a flat surface, has a significant effect on the ease of harvesting film. The lipid to protein ratio and source and form of the peanut product used in film preparation affects film formation.

Work continued on the development of a low-fat beverage. The effect of two different processing protocols on particle size distribution were studied. Beverages with little or no chalky mouthfeel can be produced when partially-defatted roast peanuts are milled and wet-blended, and slurries are filtered through a 34 μm screen before being pasteurized at 77 to 82°C and homogenized. Experiments were done to define the biochemical mechanism used by a bacterium (*Flavobacterium aurantiacum*) to degrade aflatoxin B_1. Ethylenediaminetetraacetic acid (EDTA) enhances the degradation process.

IMPACTS

The impact of technology transfer to enhance the well-being of small farmers in the Huay-Bong-Nua and Kud-Jub village areas in Thailand continues to be evident. The initiation of women cooperator groups who will process and market peanut products in two rural areas in Leyte, Philippines has had a positive impact on the families involved in this project.

A workshop on "International Training: Quality Evaluation and Utilization of Food Legumes", conducted by project members at Kasetsart University, will increase the level of awareness and knowledge of a wide range of peanut utilization topics on an international scale. The ability of the Kasetsart University team to offer this workshop, in terms of level of expertise and available equipment and instrumentation, has been made possible in large part by the Peanut CRSP. Such workshops will serve to plant the seed of knowledge which will lead to increased utilization of peanut in the Southeast Asian region and as well as in other countries in other global regions.

GOALS

A goal of the project is to enhance the capabilities of scientists, technicians and students at KU, KKU, UPLB, ViSCA and UGA. In time, these scientists will assist in disseminating information on peanut utilization in its broadest sense to research scientists, teachers, government and industry personnel and laypersons on a global scale but particularly in Southeast Asia. The education and training made possible through the project will enhance possibilities to improve the nutritional well-being of the consumer. At the same time, the economic well-being of the peanut farmer, the peanut processor and the peanut marketer will also be improved.

OBJECTIVES (1994-95)

A. <u>Kasetsart University</u>

1. Improve Quality of Traditional Peanut Products
   a. Continue to develop products containing peanut tofu by mixing with meats and vegetables. Shelf-life and consumer acceptance of this product will be evaluated.
2. Utilization of Peanut Flour
   a. Research will continue to investigate uses of texturized protein from defatted peanut flour.
3. Uses of Undersized Peanuts
   a. Research to develop a fermented product similar to sweet black soy sauce from good quality undersized peanut will be continued.
   b. Undersized peanuts will be used as a source of protein and energy to fortified ready-to-eat breakfast cereal.
4. Uses of Peanut Hulls
   a. Research on utilization of peanut hulls as packaging material will be continued.
5. Technology Transfer
   a. Cooperate with the Department of Agriculture Extension, Ministry of Agriculture to transfer technology of ground roasted peanut and peanut toffee processing to housewives in Udorn-thanee province.
6. Organize Workshop
   a. Cooperate with the Department of Technical and Economic Cooperation, Office of Prime Minister to offer a training course on quality evaluation and utilization of legumes to train participants from Indo-China countries and other countries under Thai-AID projects. The training period will be 1 month for 20 participants from those countries.

7. Postharvest Handling System
   a. Socioeconomic data collected in FY 94 in the rain-fed area of Northeastern region will be analyzed.
   b. Change in quality, with emphasis on aflatoxin contamination, of peanuts as they move through some selected postharvest handling and commercial trade channels will be monitored. Sample collection will be coordinated by Khon Kaen University and analyzed for aflatoxin at Kasetsart University.

B. University of The Philippines at Los Baños

1. Development of Peanut Products
   a. The use of peanuts and peanut by-products in snack foods, spreads, imitation cheese and comminuted meats will continue.
2. Aflatoxin Control
   a. Studies on application of compounds produced by *Cladosporium fulvum* which inhibit the growth of aflatoxigenic aspergilli will be done.
   b. Selected commercial peanut products produced in The Philippines will be monitored.
3. Industry Assistance
   a. Technical assistance to small scale industries for the purpose of improving quality and safety (aflatoxin contamination) of peanut products will continue.
   b. Assessment of processing quality of various peanut cultivars will be done.

C. Visayas State College of Agriculture

Supplementary funds will be used to conduct a research project on quality enhancement and technology transfer of products containing peanut, mungbean and cassava flour.

D. University of Georgia

1. Flavor Quality of Peanuts and Peanut Products
   a. Research will continue on the development of prediction equations for evaluating flavor quality of processed peanuts.
   b. Work on the flavor profiles of defatted peanuts will continue.
2. Alternate Uses of Peanuts
   a. The feasibility of developing a peanut spread from defatted peanuts using non-caloric fat replacers will be investigated.
   b. Optimization of conditions for solid-state fermentation of defatted peanut for the purpose of developing an ingredient for extruded snack products will be done.
3. Postharvest Handling System
   a. Data collected from the survey conducted in FY 94 will be analyzed. Plans will be developed to collect data for monitoring change in quality of peanuts at various points in the postharvest system from the field to distribution and marketing steps.
   b. Physical and mechanical properties of selected Thai peanut cultivars related to handling, storage and processing will be determined.

ORGANIZATION AND PERSONNEL

A. Kasetsart University (KU)

Department of Product Development

Dr. Penkwan Chompreeda, Principal Investigator
Mr. Vichai Haruthaithanasan, Co-Investigator
Dr. Chintana Oupadissakoon, Co-Investigator
Mr. Paiboon Thamratuwasik, Co-Investigator
Mr. Sombat Khotaveewattana, Co-Investigator
Ms. Wimolsiri Thanavisutha, Graduate Student (M.S.)
Ms. Witida Chantrapornchai, Research Assistant
Ms. Supang Reungchay, Graduate Student (M.S)
Mr. Panuwat Suppakul, Graduate Student (M.S.)

Ms. Nantana Chutimantanon, Laboratory Technician
Ms. Sathit Litlertchai, Research Technician (assisting, no support)
Ms. Kasina Boonsaeng, Research Technician (assisting, no support)
Ms. Chantana Kopayakkin, Research Technician (assisting, no support)
Mr. Chavalit Vitayanartpisal, Research Technician (assisting, no support)
Ms. Wasana Chaijaroon, Typist (assisting, no support)
Ms. Bunkerd Walkreu, Secretary (assisting, no support)

Department of Food Science and Technology, Cheingmai University
Dr. Narin Thongsiri, Cooperator

Department of Food Agricultural Engineering, Khon Kaen University
Dr. Winit Chinsuwan, Cooperator

B. University of the Philippines - Los Baños (UPLB)

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Dr. Virgilio V. Garcia, Principal Investigator
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Ms. Christia V. Almario, Research Associate
Ms. Ma. Elizabeth L. Leron, Research Associate
Ms. Arlene M. Villena, Research Associate
Mr. Raul D. Divina, Graduate Student (Ph.D.)
Mr. Reginaldo C. Calinga, Undergraduate Student
Ms. Julieta Mirabolo, Undergraduate Student

C. Visayas State College of Agriculture (ViSCA)

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Dr. Lutgarda S. Palomar, Principal Investigator
Dr. Lemuel M. Diamante, Co-Investigator
Dr. Julieta R. Roa, Co-Investigator (Socio-Ecomomics)
Dr. Remberto A. Patindol, Co-Investigator (statistics)

D. University of Georgia

Center of Food Safety and Quality Enhancement

Department of Food Science and Technology
Dr. Larry R. Beuchat, Principal Investigator
Dr. Manjeet S. Chinnan, Co-Investigator
Dr. Robert E. Brackett, Co-Investigator
Dr. Anna V. A. Resurreccion, Co-Investigator
Dr. Philip E. Koehler, Collaborator
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Dr. V. M. Balasubramaniam, Post Doctoral Associate (partial support)
Dr. Isam Hashim, Post Doctoral Associate (assisting , no support)
Mr. Witoon Prinyawiwatkul, Thai Graduate Student (M.S.)
Mr. Anuvat Jangchud, Thai Graduate Student (Ph.D)
Ms. Grace Divino, Filipino Graduate Student (M.S.)
Mr. Agustin Ramos, Filipino Graduate Student (M.S.)
Ms. Doris D'Souza, Graduate Student (Ph.D)

Mr. Alan Hathcox, Graduate Student (M.S.) (partial support)
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Ms. Brenda Nail, Agricultural Research Coordinator I (assisting, no support)
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Mr. Lary Hitchcock, Research Technician III (assisting, no support)
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Ms. Brenda Banister, Accounting Assistant (assisting, no support)

ACCOMPLISHMENTS IN DETAIL

RESEARCH

A. Accomplishments at Kasetsart University (KU)

1. Development of a Breakfast Bar Using Undersized Peanut as Source of Protein

A consumer survey was conducted to identify a desirable prototype product of a breakfast bar. Results from 100 consumers indicated that they preferred a rectangular shaped bar weighing 50-100 g. Consumers preferred a product containing pineapple flavor, crispy in texture and moderately sweet. Linear programming was employed to formulate a bar to meet the RDA requirement for protein, iron, vitamin A, thiamin, riboflavin and niacin. Six formulas were prepared and quality characteristics were measured. Statistical analyses were employed to optimize the bars. The optimum formula of a breakfast bar contained 15% crispy rice, 20% roasted peanut, 5% dry banana, 18% sun flower seed, 3.2% glucose syrup, 15% pineapple jam, 5% dry milk, 4.3% honey, 5% coconut sugar, 8.5% water, 0.5% pineapple flavor and 0.5% salt. Ingredients were mixed, molded into 2.5 x 3 x 1.5 cm forms and packed in aluminum foil. Process optimization, shelf-life study and consumer acceptance will be determined.

2. Development of a Snack from Texturized Peanut Protein

Research was conducted to further investigate the development of a snack product from Texturized Peanut Protein (TPP). Snack products were the target since TPP contains more than 50% protein. This study also aimed to develop a more nutritious product for Thai consumers. Results of a survey indicated that consumers (120 Thai adults) preferred snacks containing barbecue flavor that were moderately sweet and salty, and with crispiness and light color. The optimum formula contained 82.3% defatted peanut flour, 14.0% sugar, 2.5% salt and 1.2%. The process of preparation of the product is shown in the Fig. 1. The snack is yellow-brown in color with a water activity of 0.34. The compressive strength of the snack is 13.1 Newtons. The product contained 4.0% moisture, 46.6% protein, 4.0% fat, 6.3% fiber, 4.7% ash and 34.4% carbohydrate. The total microbial count and yeast and molds were less than 10 CFU/g. Acceptance tests showed that consumers like the snack moderately.

Defatted peanut flour + Sugar + Salt + Barbecue Flavor
|
Add water (35%)
|
Mix, knead
|
Weigh 15 g each
|
Flatten by hand
|
Texturize in village scale texturizer at 165°C
|
Press for 10 sec
|
TPP snack
|
Slice (0.5 cm thick)
|
Dry at 65°C for 30 min
|
Package in aluminum foil bag

Fig. 1. Flow diagram for preparing TPP snack.

3. Effect of Substrate on Efficiency of Peanut Tempeh Starter

Peanut tempeh has potential as a protein source for Thai consumers. Several products prepared from peanut tempeh were introduced to consumers and acceptability is promising. This study was conducted to investigate the effect of substrates used in preparing starters on quality of peanut tempeh. Seven formulations of substrate were prepared, innoculated with a pure culture of *Rhizopus oligosporus* and incubated at ambient temperature (30°C) until full sporulation was achieved. Starters were dried at 50°C for 4 hr, and then used to prepare peanut tempeh. Good quality peanut tempehs were obtained using three different starters: 1) rice flour (60%) + rice bran (6.7%) + water (33.3%), 2) rice flour (53.4%) + ground deskinned raw peanut (13.3%) and 3) rice flour (40%) + ground deskinned raw peanut (26.7%) + water (33.3%). The first formula was the least expensive and therefore was used in scale up production to compare with a commercial starter from The Tempeh Lab. The starter was sent to housewives at Kud-Jub District, Udorn-thanee Province to test their acceptability for uses in preparing peanut tempeh. Preliminary results indicate that of housewives were very comfortable in using the starter. Their peanut tempeh was of good quality and not contaminated with other microorganisms. In addition, housewives prepared several local products from fresh and dried peanut tempeh. This study will continue as part of the technology transfer effort.

4. Development of Mosquito Repelling Sticks using Peanut Hulls

A consumer survey was conducted to identify a desirable mosquito repelling stick prototype. Results from 100 consumers indicated that they preferred a mosquito repelling stick forming into a coil shape. Consumers preferred a product containing citrus aroma. Mixture design experiments were employed to formulate products to meet the concept. Sticks contained 40% peanut hulls. Particle size of all ingredients was between 250 - 500 μm to form a product with low rate of combustion. A mixture of ingredients (peanut hulls, lemon grass, citrus leaves, orange peels, tapioca flour and filler) at 47.5% was combined with 52.5% water extruded into sticks and dried at 60°C for 80 min. Process optimization, shelf-life studies and consumer acceptance will be determined.

5. Improving the design of a sheller for a large seeded cultivar (Khon Kaen 60-1).

A peanut cultivar (Khon Kaen 60-1) has been successfully bred and adapted for Thailand. The kernels of this cultivar are substantially larger than those of Tainan-9, a commonly grown cultivar. However, the new cultivar has not been widely adopted by farmers due to many practical problems. Breeders have tried to promote this cultivar in specific areas. A feasibility study for producing the Khon Kaen 60-1 cultivar for commercial use has also been conducted. With the larger size of the new cultivar, the existing sheller developed at KKU can not be used. Therefore, to support the promotion program for the Khon Kaen 60-1, design criteria for modification of the rubber tire sheller were developed. The factors studied were as follows:

a. *Concave rod spacing:* Six spacings ranging from 0.5 mm smaller than the average maximum width of kernel 0.5, 1.0, 1.5, 2.0 and 2.5 mm larger than the average maximum width of kernel.
b. *Clearance between rubber tire and concave*: Four spacings at 0.5, 3.5, 6.5 and 9.5 mm larger than the average maximum width of kernel.
c. *Shelling speed*: Five speeds selected were 3.86, 5.41, 6.95, 8.50 and 10.04 m/s.
d. *Feed rate*: Four rates chosen were 250, 300, 350 and 400 kg/h.
e. *Pod moisture*: Four levels selected were 5.3, 9.7, 2.0 and 15.6% (wb).
f. *Shelling concave*: Having rotating concave rods perpendicular or parallel to rotating direction of the rubber tire were also compared.

Shelling efficiency and percentage of broken kernel were used as indicators for performances. Recommendations for modification of the Motorized Rubber Tire Sheller for Khon Kaen 60-1 were as follows: The shelling concave with rotating concave rods perpendicular to the rotating direction of the rubber tire is recommended, with clearance between rods 0.5 to 1.0 mm larger than the average maximum width of the kernel. The clearance between the rubber tire and concave should be 6.5 to 9.5 mm larger than the average maximum width of kernel. The shelling speed should not exceed 8.5 m/s shelling peanuts with moisture content of 12.0 to 15.6% (wb). For peanuts with moisture less than 10%, the speed should be less than 3.86 m/s. Feed rate should be less thatn 400 kg/h.

B. Accomplishments at the University of the Philippines at Los Baños

1. Utilization of Peanut in Foods

a. Extended meatloaf

The suitability of peanut as an extender of ground pork and beef in meatloaf was evaluated. Boiled peanut, defatted peanut and peanut residue were incorporated into a meatloaf formula consisting of the following ingredients (in g): lean beef (50), lean pork (8.8), back fat (25), salt (4), sugar (2), pepper (0.2), nutmeg (0.03), powdered garlic (0.3), praque power (0.35), monsodium glutamate (0.25), fosfat (1), ascorbic acid (0.12), skimmilk (12.6), ice water (62) and peanut, defatted peanut or peanut residue (83.3). Frozen beef, pork and back fat were passed thru a meat grinder. The other ingredients were added and the mixture was ground again. The mixture was then dispensed in loaf pans and baked at 80°C for h and further at 120°C until the internal temperature reached 105°C or the loaf became golden brown. The loaves were cooled, removed from pans and stored in the freezer.

The peanut extended meatloaves were subjected to sensory evaluation to determine differences in color, aroma, texture and general acceptability. Products were significantly different from each other in meat aroma, peanut aroma, meat flavor, peanut flavor and general acceptability and not significantly different in color, off flavor and

texture. The formula containing 50% peanut residue was most acceptable, having the weakest peanut flavor and the least perceptible off flavor. The best formulation containing peanut was compared with a 100% meat product extended samples were significantly different in terms of color, aroma, meat aroma, meat flavor, texture and general acceptability and not significantly different in terms of flavor and off-flavor.

The proximate composition was determined. The high protein and fat contents make peanut residue a valuable ingredient to various food preparations instead of discarding it after peanut milk extraction. Moisture content of the residue was adjusted to about 10% by oven drying at 55 to 60°C for 5 hours after extraction. The functional properties of peanut residue are favorable for wide applicability.

Improvements on the formulation and processing of peanut-meatloaf were done to simulate commercial counterparts. To the original formulation, 25 g of cornstarch was added and water was reduced to 25 ml. The mixture was packed in 211 x 300 cans and processed at 121°C for 30 min. The resulting product was compared to two commercially available meatloaves for sensory attributes, proximate composition and other physical properties (color and hardness). Results showed that peanut-substituted meatloaf is equally acceptable to commercial meatloaves with similar nutritional value and physical characteristics.

b. Peanut nuggets

A peanut nugget were developed using the following formula (g): peanut residue (175), salt (6.1), sugar (2), monosodium glutamate (0.25), black pepper (1), garlic powder (0.5), skimmilk (15), cornstarch (25), flavoring (0.1), water (25) and colorant (yellow) (0.5). Ingredients were combined, mixed, shaped into cubes (1 inch) and rolled in bread crumbs. The nuggets were then deep fried until golden brown and placed on paper towels to remove excess fat. Proximate analysis and sensory analysis was done. The addition of artificial flavoring did not significantly affect the color, aroma, flavor, texture or general acceptability. This product is suitable as a snack or served with sauce.

c. Peanut cracker

Peanut crackers were made using peanut butter, flour, starch, baking powder, salt, sugar, monosodium glutamate, vegetable oil and water. Peanut varieties from the Institute of Plant Breeding (Pn 2, Pn 4, Pn 10, and IPB. 87-21-26) as well as four others purchased from Divisoria were processed into peanut butter. Peanuts were roasted at 350°F for 40 minutes. An emulsifier at 1.6% level (Dimodan PV from Marsman) was added while the peanuts were still hot, then sugar, salt, and vegetable oil were added. The mixture was ground three times or until smooth. The product was placed in sterile bottles and evaluated after 30, 60, and 90 days at room temperature. Moisture content, crude fat content and total sugars of the peanut samples were determined. Color measurements were made using a Minolta CR-200 Series Chroma Meter on the peanut butter samples. Oil separation, free fatty acid, and peroxide value were monitored after every storage interval. The type of flour, the starch to flour ratio, levels of baking powder, and ratio of oil to sugar solution for spraying were varied. Sensory evaluation was conducted using quality scoring for appearance, flavor, hardness, crunchiness, and general acceptability. Data were analyzed using ANOVA and DNMRT.

The size and weight of the peanut kernels were important factors affecting the color of the peanut butter. Chemical analysis revealed that the crude fat content of the peanuts obtained from Divisoria was relatively higher than that of the IPB samples. Storage studies showed that oil separation is very minimal in peanut butters made from the IPB varieties as compared with peanut butters made with Divisoria peanuts. The peanut butter samples were more prone to hydrolytic rancidity than to autoxidation.

Various formulations of peanut cracker had high acceptability. The judges did not detect significant differences among the three types of flour used, namely bread, cake, and all-purpose. Sensory evaluation revealed that the optimum amount of vegetable oil and sugar solution needed for spraying is 7.7 - 15.4% and 84.6 - 92.3%, respectively. The optimum amount of baking powder was 13.7%. At this level, a highly acceptable peanut cracker could be produced using 70 - 80% and 20 - 30% level of flour and starch, respectively.

2. Microbiological Control of Aflatoxin

Crude extract of the metabolite of a mold exhibiting inhibitory activity toward aflatoxin production was obtained by adjusting the pH of the filtered culture broth to 3.5-4.5 and adding 2% activated charcoal, followed by extraction with methanol or acetone and drying in a rotary evaporator at 40°C. Further purification was done with n-hexane. The extract was purified by gel filtration in a Sephadex LH-20 and then subjected to HPLC for detecting the active compound. Each fraction collected in HPLC was tested for biological activity with the active fractions concentrated and dried for spectroscopic studies. Ultraviolet (UV) absorption spectrum, infrared (IR) spectrum, nuclear magnetic resonance (H NMR) spectrum and mass (MS) spectra were used to analyze samples. Gel chromatography and HPLC peak fractions were tested for biological activity against *A. parasiticus*, *A. flavus* and *Alternaria kikuchiana* using the paper disc technique.

Concentrated acetone and methanol extracts yielded a waxy or oily red colored residue of about 0.66 g/l of culture broth. Further extraction with n-hexane resulted to a reddish residue of about 0.44 g/l of culture broth. Microassays of crude extracts from gel filtration showed that the inhibitory metabolite is a compound closely associated to the red pigment. The pooled active fractions yielded a colorless cystalline residue of about 80 mg/l of culture broth. The final purification process by HPLC yielded a residue of about 6.2 mg/l of culture broth. Based on spectroscopic studies of the active metabolite, the compound appeared to have 274 as its molecular weight and its tentative formula is $C_{10}H_{20}O_2$.

A stable form of the metabolite was prepared by freeze drying the samples. The completely dried culture broth was difficult to handle because it absorbs moisture once exposed at room temperature. A powdered form of the metabolite was prepared by freeze drying the culture broth until 87% of the broth was evaporated, mixing (cornstarch:freeze dried culture broth ratios) with cornstarch at 1:1, 1:3 and 1:5 and drying at 55°C. Preliminary application of the powder indicates that the 1:3 proportion was most effective to use. Potency and stability tests were done using *A. parasiticus* as the test organism. Its effectiveness in controlling growth of *A. parasiticus* in peanuts was also determined.

Results from the microassay test showed that the powdered form is more effective compared to the culture broth per se and that it is stable up to 3 months storage under refrigeration temperature. Application of the powder to peanuts was not effective due to its inability to completely coat the peanuts. Use of culture broth per se on peanuts was still more effective but only up to 2 weeks. This may be due to the decrease in potency of the metabolite or to the added nutrients which promoted growth of *A. parasiticus*.

C. Accomplishments at Visayas State College of Agriculture

1. Peanut Product Quality Enhancement

a. Supplementation of Lady Finger to Enhance Nutritional Value

Since Lady Finger made from cassava has a lower crude protein content, supplementation with peanut was done to increase its protein content and overall value. Peanut was identified as one of the acceptable enrichment/flavoring ingredients from a market survey and market and product testing done in FY95. Taste panels showed no significant differences in mean scores between control and enriched products. However, since Lady Finger was the first pilot produce tested, cooperators resisted in preparing and marketing it as peanut-enriched product since the original recipe was already acceptable and supplementation would increase cost. This situation and a problem with breakage of Lady Finger gave birth to the formulation of Lady Buttons.

Experiments revealed the potential for adding about 20% peanut to produce products with acceptable sensory qualities and higher protein content. Reformation of with the Peanut-ChoCass product, already manufactured by a ViSCA cooperator, was also done. Increasing the content of peanut and cocoa powder in this product did not significantly improve its sensory qualities, except for color (Table 1). The scores for color increased with an increase in the level of cocoa. Results revealed the possibility of reducing the amount of cocoa in the formulation and still producing acceptable products but a reduced production cost.

b. Optimization of Flour/Starch Content of Lady Finger

The formulation optimization process for Lady Finger first required a decision on the variables to be studied. A simplex centroid design and mixture response surface methodology (RSM) were used to determine the influence

blends of the three variables (wheat flour, cassava flour and cassava starch) on sensory qualities. Products were subjected to sensory evaluation at ViSCA and consumer testing. The data were analyzed, and regression coefficients were (Fig. 2) used to generate contour plots which were later overlaid to obtain the optimum regions of formulation.

The shaded regions in Fig. 2a and Fig. 2b represent values for consumer acceptance for a particular attribute of the Lady Finger product corresponding to scores of ≥7.0 (like moderately). For most sensory qualities, the optimum regions were toward the peripheries of the triangle. This indicates similar levels of suitability of the three components as individual ingredients or in combination with the others. Consumer acceptance for texture appeared to be the limiting factor (Fig. 2a) during the optimization procedure. This was the only factor which gave only one treatment with a mean score of about 7.0 (Table 2). Only a very small region satisfied the requirements of scores ≥7. 0 (Fig. 3). No more than about 2.0% of cassava flour can be added to the formulation to produce a flavor of 7.0 or better. However, a bigger region satisfied the requirements of scores ≥6.5 with even 100% cassava flour (Fig. 3b).

Table 1. Mean taste panel scores of chocass containing various levels of peanuts and cocoa.

Ingredient		Sensory Qualities			
Peanuts (g)	Cocoa (g)	Color	Texture	Flavor	Overall Accept.
0	0	6.0b	6.9a	6.7a	6.6a
0	15	7.2a	7.3a	7.3a	7.3a
0	30	7.5a	7.1a	6.9a	7.1a
30	0	6.0b	6.9a	6.8a	6.8a
30	15	7.4a	7.2a	7.3a	7.3a
30	30	7.4a	6.8a	6.7a	6.8a
60	0	5.9b	6.6a	6.6a	6.6a
60	15	7.2a	7.0a	7.1a	7.1a
60	30	7.2a	7.0a	7.1a	7.1a

Table 2. Mean taste panel scores of Lady Finger containing combinations of wheat flour (WF), cassava flour (CF) and cassava starch (CS) used during the optimization studies*.

WF	CF	CS	Sensory Attributes			
%			Color	Texture	Flavor	Gen. Accept.
100	0	0	7.2	6.8ab	7.1a	7.1a
0	100	0	6.7	6.5abc	6.4bc	6.4bc
0	0	100	6.3	6.2c	6.3c	6.2c
50	50	0	7.2	6.8ab	7.1a	7.2a
0	50	50	6.5	6.4bc	6.8ab	7.0a
50	0	50	6.9	7.0a	7.0a	7.2a
33	33	33	7.1	6.7ab	7.1a	6.9a
67	16.5	16.5	6.7	6.6abc	6.6abc	6.7ab
16.5	67	16.5	7.1	6.9ab	6.6abc	6.8ab
16.5	16.5	67	7.6	6.7abc	6.8ab	6.9a

*Means followed by similar letter are not significantly different at 5% level.

c. Packaging and Shelf life Studies on Baked Rootcrop-Peanut Products

Several new baked products containing rootcrops have been developed. Three of these products are the Lady Finger, Lady Buttons and Peanut-Sweetpotato (PS) Cookies. Lady Finger traditionally contains cassava flour and is a baked product with an elongated shape. Lady Button is a variation of Lady Finger in that it contains crushed peanuts and has a rounded shape, while PS Cookies is a baked product containing sweet potato flakes and crushed

peanuts. These products have good potentials for commercialization; however, there is a need to package the products properly in order that they reach the consumers in good condition.

Selection of the proper package of any product requires knowledge of the relevant mechanisms of deterioration and, particularly, the effect of water activity and temperature on deterioration. Preliminary studies showed that freshly made products always had sensory scores of about 7. The moisture contents of representative packed samples of various formulations were determined by oven drying at 105°C for at least 15 h. The moisture content of the sample which got a sensory score that was significantly lower than the control sample was considered as the critical (unacceptable high) moisture content of the product.

i. *Shelf life prediction*
The absorption isotherms of Lady Finger, Lady Buttons and PS Cookies at 30°C were determined. The experimental isotherms were fitted with a linear equation to get the slope and intercept values needed for packaging calculations. The shelf

life of the three products were predicted for accelerated storage conditions (38°C and 90% RH) packaged in density polyethylene (LPDE), high density polyethylene (HDPE) and polypropylene (pp), all commonly available packaging materials.

ii. *Accelerated shelf life testing*
Based on the predicted shelf lives of the products, samples of Lady Finger, Lady Buttons and PS Cookies were packaged in LDPE, HDPE and PP. Products were placed in airtight containers at 90% RH in an incubator at 38±1°C for several days before being evaluated for sensory qualities.

Results of the sensory evaluation of Lady Finger for critical moisture content are shown in Table 3. Six exposure time durations ranging from 5 to 240 min were studied. The initial moisture content of the product increased from 1.13% (dry basis) to 8.26%. Based on the mean sensory scores for loss of texture, the critical moisture content of Lady Finger is about 2.28%, since the sensory score of 6.26 of this sample is significantly lower than the score of 7.53 for the control sample and the higher bound value (6.86) was below 7.

Table 4 shows the results of the sensory evaluation of Lady Buttons for critical moisture content determination. Five exposure time durations ranging from 1.5 to 20 min were studied. The initial moisture content of the product increased from 0.93% to 3.43%. The critical moisture content of Lady Buttons is not as well defined as for Lady Finger. For Lady Buttons the critical moisture content is around 2.86%, since samples with this level gave a texture score of 6.80, which is below the cut-off score of 7.

Table 5 shows the results of the sensory evaluation of PS Cookies for critical moisture content determination. There was at total of six exposure time durations ranging from 15 to 180 min. The initial moisture content of the product increased from 1.15% to 4.99%. The critical moisture content is around 3.56%, since the sensory score of 6.40 (6.81) is still below 7.

Fig. 4 shows the adsorption isotherms (regression lines) at 30°C. The fitted linear equations for the three products are as follows:

Lady Finger: $Me = 1.92856 + 13.86988\ Aw$ (r^2=0.981)
Lady Buttons: $8.77264\ Aw$ (r^2=0.984)
PS Cookies: $Me = 0.90862 + 7.77171\ Aw$ (r^2=0.987)

where: Me = equilibrium moisture content (% dry basis) and Aw = water activity (dimensionless)

These equations have high coefficients of determination (r^2), which indicates a high degree of fit of experimental and predicted data. It is also worth noting that the lower the critical moisture content of the sample, the higher intercept and slope of the regression line. Results indicate the Lady Finger is more moisture sensitive in terms of moisture uptake compared to Lady Buttons and PS Cookies. Lady Buttons and PS Cookies contain crushed peanut which gives them higher oil content than Lady Finger.

Table 3. Moisture content and mean sensory scores (20 panelists) for loss of texture of Lady Finger using a9-point Hedonic scale. Samples were exposed at various time durations at 100% relative humidityand ambient temperature.

Exposure time (min)	Moisture content (% dry basis)	Mean score	95% Confid. bound
0 (Control)	1.13	7.53	±0.37
5	1.53	7.11	±0.45
15	2.28	6.26	±0.60
45	2.99	5.74	±0.71
75	5.00	5.74	±0.55
135	5.86	4.79	±0.90
240	8.26	4.11	±0.92

Table 4. Moisture content and mean sensory scores (20 panelists) for loss of texture of Lady Buttons using a 9-point Hedonic scale. Samples were exposed at various time durations at 100% relative humidity and ambient temperature.

Exposure time (min)	Moisture content (% dry basis)	Mean score	95% Confid. bound
0 (Control)	0.93	7.35	±0.32
1.5	2.28	6.95	±0.23
3.0	2.42	7.05	±0.30
5.0	2.56	7.00	±0.42
10.0	2.86	6.80	±0.41
20.0	3.43	7.05	±0.46

Table 5. Moisture content and mean sensory scores (20 panelists) for loss of texture of PS cookies using a 9-point Hedonic scale. Samples were exposed at various time durations at 100% relative humidity and ambient temperature.

Exposure time (min)	Moisture content (% dry basis)	Mean score	95% Confid. bound
0 (Control)	1.15	7.50	±0.32
15	2.63	7.40	±0.23
30	2.99	7.25	±0.30
60	3.32	6.80	±0.42
90	3.56	6.40	±0.41
120	3.75	6.15	±0.46
180	4.99	5.50	±0.44

The calculated BET monolayer moisture contents at 30°C were 2.99% for Lady Button, 4.86% for Lady Finger and 2.38% for PS Cookies. These moisture contents were compared with the critical moisture contents of the same set of products obtained earlier. Results showed that only Lady Buttons have similar monolayer and critical moisture contents of about 3% dry basis. The critical moisture content of Lady Finger was lower than the monolayer moisture content while, for PS Cookies, the opposite was observed. Thus, Lady Buttons are predicted to be more stable compared with Lady Finger and PS Cookies.

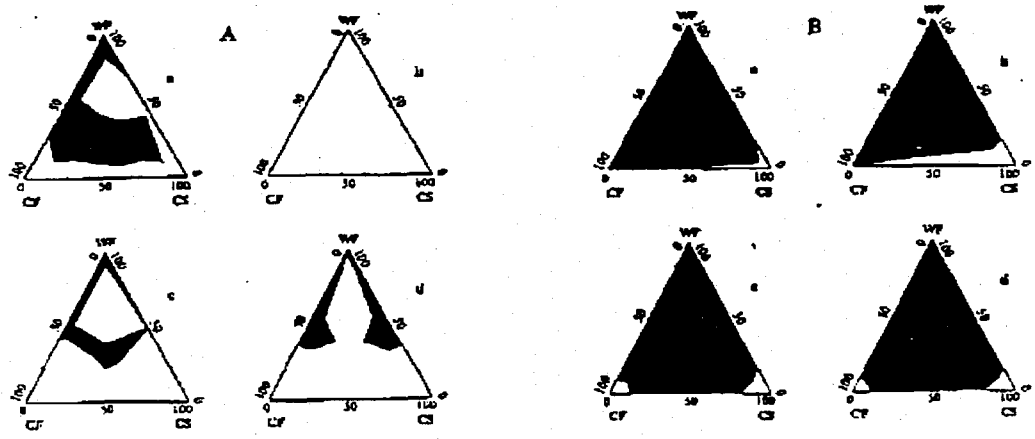

Figure 2. (A) -Contour plots for quality blends containing wheat flour (WF), cassava flour (CF), and cassava starch (CS) in Lady Finger for (a) color, (b) texture (c) flavor, and (d) overall acceptability. The shaded regions represent the acceptability scores of ≥ 7.0;

(B) -Contour maps for quality parameters of parameters of blends containing wheat flour (WF), and cassava starch (CS) in Lady Finger for (a) color (b) texture (c) flavor, and (d) overall acceptability. The shaded regions represent the acceptability scores of ≥ 6.5.

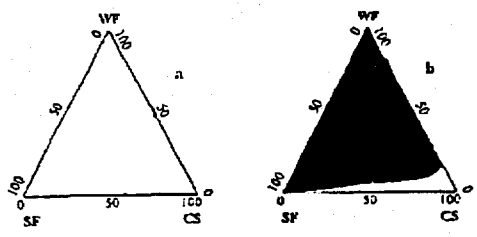

Figure 3. Superimposed regions after overlaying the optimum regions of the different quality parameters at scores ≥ 7.0 and ≥ 6.5 (b).

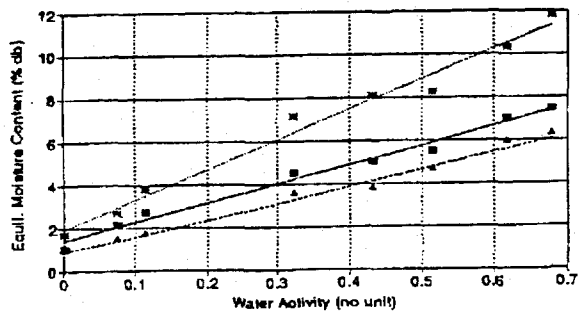

Figure 4. Absorption isotherms for Lady Buttons (■), Lady Fingers (*) and Peanut-Sweet potato (▲) cookies at 30°C

Using appropriate equations, the predicted shelf life of Lady Finger packaged in LDPE, HDPE and PP at accelerated conditions were calculated (Table 6). Confirmatory studies were conducted at accelerated conditions of 38°C and 90% RH for the same product. Shelf life was estimated by sensory evaluation. The storage time of the sample that got a mean sensory score of less than 7 was considered as the shelf life of the product. The actual shelf life of stored Lady Finger stored in various packaging materials is shown in Table 6. There was satisfactory agreement for the predicted and actual shelf life for Lady Finger. Similar results were also obtained for Lady Buttons as shown in Table 7. Prediction of shelf life for PS Cookies was also satisfactory, except for samples stored in LDPE, where the actual shelf life was underestimated by half a day (Table 8). Results suggest that a packaging equation can be used to predict the shelf life of stored peanut-supplemented products.

By rearranging equations packaging requirements for Lady Finger, Lady Buttons and PS Cookies may be determined. The calculated package thickness is based on accelerated storage conditions of 38°C and 90% RH), a multiplication factor needs to be introduced. Shelf life of dry snack foods stored at ambient conditions is 7-10 times that at accelerated conditions. Taking a value of 8, the desired shelf life of about 1 month (32 days) at ambient conditions will be approximately 4 days at accelerated conditions. Using the same packaging materials, bag dimensions and sample characteristics, the thickness of the bags were obtained (Table 9). Lady Finger requires packaging in 0.003626 inch thick pp to achieve a shelf life of about one month at ambient temperature. Lady Buttons need to be packaged in only a 0.002095 inch thick of pp bag to achieve the same shelf life, while for PS Cookies, the thickness of the pp bag is 0.0001222 inch thick. Based on commercially available pp bags in Metro Cebu (Table 10), the thickness of the bags for Lady Finger, Lady Buttons and PS Cookies will be 0.00374, 0.00216 and 0.00118 inch, respectively.

Table 6. Predicted and actual shelf lives of Lady Finger at accelerated conditions of 38°C and 90% RH using different packaging materials.

Packaging Material	Thickness (inch)	WVTR (g water /{m^2.day})	Shelf Life (day) Predicted	Actual
Low density polyethylene (LDPE)	0.00246	11.6	1.95	2.0
High density polyethylene (HDPE)	0.00098	3.8	1.34	1.5
Polypropylene (PP)	0.00197	8.9	2.66	2.5

Table 7. Predicted and actual shelf lives of Lady Buttons at accelerated conditions of 38°C and 90% RH using different packaging materials.

Packaging Material	Thickness (inch)	WVTR (g water /{m^2.day})	Shelf Life (day) Predicted	Actual
Low density polyethylene (LDPE)	0.00246	11.6	3.04	3.0
High density polyethylene(HDPE)	0.00098	3.8	3.70	4.0
Polypropylene (PP)	0.00197	8.9	3.17	3.0

Table 9. Packaging material thickness needed for a shelf life of 32 days for Lady Finger, Lady Buttons and PS cookies at ambient conditions (Assumptions: M_1 =1.20% d.b.: Weight of product = 35 g; Package dimensions: 0.15m x 0.125m).

Packaging material	Packaging material thickness (inch) Lady finger	Lady buttons	PS cookies
Low density polyethylene	0.004726	0.002726	0.001592
High density polyethylene	0.001548	0.000894	0.000521
Polypropylene (ordinary)	0.003626	0.002095	0.001222

Table 10. Packaging materials and their corresponding thicknesses that are commercially available in Metro Cebu.

Packaging Material	Thickness (inch)
Polypropylene	0.00098
	0.00118
	0.00197
	0.00216
	0.00374
Low density polyethylene	0.00246
High density polyethylene	0.00030
	0.00098

2. Technology Transfer and Commercialization Strategies

a. The Approach: Interdisciplinary and Integrated

Project team members, consisting of a food scientist, a food engineer, a socioeconomist and a statistician, facilitate the technology transfer. The members held more interactive meetings and consultations for planning, implementation and report writing while each, at times, carried out respective disciplinary tasks to contribute to the whole process. Thus, team and individual site visits were carried out. This proved to be a more effective approach in hastening the transfer process. Participative in nature, immediate feedback from both the processors

the whole process. Thus, team and individual site visits were carried out. This proved to be a more effective approach in hastening the transfer process. Participative in nature, immediate feedback from both the processors and the market made possible modifications of the product, process and operations relatively more responsive and timely.

b. Linkage Establishment and Inventories

After the establishment of linkages and consultative meetings, an inventory of skills and facilities followed. This was necessary to speed up the adoption process. Previous exposure of members to ViSCA technologies enabled them to make quick decisions as to what to produce and technology to adopt.

c. Organizational and Entrepreneurial Strengthening

i. *Entrepreneurship development trainings.*

The capacity of would-be adopters (women cooperators) is critical in the successful and sustainable transfer of innovations. Thus, in the early phase of pilot testing of product technologies with the cooperators, a program on skills development other than processing has been established. Aside from informal consultations with cooperators as part of the organizational build-up process, two 2-day sessions on entrepreneurship development training were conducted. These sessions were designed to suit the schedule of the women cooperators who are also mothers and workers, and to fit to their identified skills-training needs. The first session focused on the basics of starting a business, marketing and record keeping. Resource persons from the Department of Agricultural Economics and Agribusiness of ViSCA gave the lectures. A field visit with pilot processing cooperators in Baybay enriched that training component

d. Processing Skill Improvement

i. *Guadalupe Women Association (GWA)*

The processing center of the GWA was built on the government lot of Guadalupe, Baybay, Leyte with financial assistance from ViSCA GTZ Program. The group's contribution was labor and other supplies especially gravel and sand. An oven was loaned by ViSCA while processing utensils, tools and small equipment were purchased by the group using their initial capital and profit from catering activities.

ii. *URIVIC*

The processing center of URIVIC is a kitchen belonging to Philippine National Red Cross (PNRC). The kitchen is equipped with a stove, refrigerator, freezer, plastic sealer, weighing scale and other kitchen utensils and tools, since these were used earlier by a meat processing group. Before skill training was done, basic baking equipment was added to the facilities. PNRC purchased some of these items, while an oven was also loaned to the group by ViSCA.

Continuous skill training was done with the two groups, either in response to feedback during the market and consumer survey or product line expansion process. The process has been continually evaluated to improved for efficiency. Raw material sourcing and processing skill was also monitored and improved. Repeated visitation was one of the strategies to enrich their processing skills.

Intragroup sharing was also encouraged with both groups. Members who were previously trained and/or have experience became the "product experts" within the group, and trained the other members to serve as support or standby processors in time of absences and demand for increased production volume for local or expanded markets.

e. Market Development

After the initial consumer surveys and product tests, the Lady Finger and Lady Buttons, and PS Cookies were produced by URIVIC and GWA, respectively. Packaged products (30-50g) were market tested locally in the

cities of Ormoc and Tacloban, and in Los Banos in Luzon and in Mindanao. The products were also introduced and sold in trade fairs, exhibits, research and development conferences and workshops.

i. *Improving product presentation*

Packaging is an important component in product selling. The groups were assisted in the design and searching out of packaging materials for the cookies. To reduce the need for a variety of packaging materials to be used for various products and, thereby, offset the minimum order requirement by the manufacturer, multi-product packaging was used by both groups. This is a critical strategy, since the groups have limited initial capital.

ii. *Market expansion.*

It is necessary to increase the volume of processed products to make the groups' operations viable and sustainable. Thus, assisting the groups in expanding their market outlet is a basic part of the technology transfer scheme.

iii. *Guadalupe Women's Association*

The GWA regularly sell various rootcrop-based products in it small village store. Since this caters to a very limited market, GWA assisted to secure a market stall and sell in the town of Baybay during fair days, on Tuesdays and Saturdays. GWA also promoted and sold the products from other ViSCA-assisted rootcrop processors. However, this activity was not sustained, since the members' processing and household activities schedule had to be given priority. Only five members of GWA can fully commit their time to the process due to other obligations. The group agreed to try other increased marketing schemes such as contacting local sellers to sell their products for a percentage share of the sales. Selling strategies are still being explored.

The PS Cookies were tested in local markets, Ormoc, (Tacloban and Los Banos). Results showed that the quality of the product needs to be stabilized. The quality characteristics which need to be looked into are the size and hardness of the peanut grains, sweetness, appearance and color. The processors, too, must be careful in sealing the packs as this affects the shelf-life of the products. Direct feedback from the market to the researchers to directly work on the suggested improvements with the processors.

iv. *URIVIC.*

The Lady Finger and Lady Buttons produced by URIVIC were also market tested in the same areas. Both the Lady Finger and the Lady Buttons have been gaining market in the outlets where they were introduced as indicated by repeat buys. The latter product had an advantage because it does not need the delicateness in handling which the Lady Finger requires.

The number of outlets of URIVIC has tripled to include a newly contacted supermarket in Tacloban. Most of its outlets are snack and ice cream houses, school and office canteens. The quality of both products have already been stabilized at the processors' level. The return on investment (ROI) averaged 30%. The critical needs which still have to be met are improvements in packaging for better presentation and handling and a cost-effective product distribution system.

v. *Sales monitoring.*

A bi-monthly summary of production showed an irregular output for both groups, figuring relatively high in months with more orders for market testing activities. This reflects the relative weakness of the marketing efforts, as the groups were still in their initial learning phase, as well as a lack of a stable supply of rootcrop flour. Efforts are being undertaken to stabilize the supply of cassava flour from a cooperative in the South to sustain market gains.

For both groups, further marketing assistance is needed to train members in the marketing process itself; that is, getting and putting to work the market feedback, seeking and opening outlets and establishing channels of distribution and promotional strategies.

d. Project Monitoring

A feedback mechanism consists of site visits and consultations, and cooperators' workshops where experiences are shared. These provide experiential learning and the essential information needed for the research optimization in the laboratory, the standardization of activities at the groups' processing centers and successful transfer of technologies in general.

D. Accomplishments at the University of Georgia

1. Development of Honey-Peanut Butter Products

In studies to increase the utilization of peanuts in value-added products for U.S. markets, a honey-peanut butter product was developed. Studies were undertaken to formulate prototypes of honey-peanut butter (HPNTB), to optimize HPNTB for consumer acceptability, and to characterize flavor, texture, and appearance of optimized HPNTB. Shelled, U.S. No. 1 medium runner type peanuts were used in the preparation of the peanut butter. A 3x3 factorial design was used to investigate the effect of honey and salt level on sensory properties of peanut butter. Formulations containing three different levels of dry honey powder and salt were used. In addition to dry honey (6, 13 and 20%) by weight of ground roasted peanuts and salt (1, 1.5 and 2.0%), a commercial stabilizer (Fix-X, Proctor & Gamble, Cincinnati, OH) was used at 1.25% by weight of ground roasted peanuts. A control peanut butter (containing no honey) was prepared using 6% corn syrup solids, 1.25% Fix-X and 1% salt by weight of ground roasted peanuts.

Consumers were recruited from a consumer database list (metropolitan Atlanta area) and from faculty, student and staff at the University of Georgia to evaluate products. Participating consumers (n = 93) were screened to be regular consumers of peanut butter (at least eat peanut butter twice a week), were between 18 and 65 years of age, and were available and willing to participate. Consumers were selected to represent a range of educational backgrounds, marital status, household incomes, and employment status. The tests were conducted in a sensory laboratory in partitioned booths. Consumers were asked to determine acceptability of color, spreadability, sweetness, saltiness, flavor, texture, and overall acceptance of peanut butter containing honey and a control (regular peanut butter) using a 9-point hedonic scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely). Consumers were asked to assess their intention to purchase test samples and the price they were willing to pay for the samples compared to the price they are now paying for peanut butter.

Three to five consumer test sessions were conducted from 9:00-11:30 a.m. and 2:00-6:00 p.m. over a period of three consecutive days. During each session, each consumer evaluated five samples, had a break for 10 min, and then evaluated five additional samples. The samples were presented to consumers in 1-oz plastic cups coded with 3-digit numbers and served in random sequential monadic order with white bread. Consumers were instructed to spread peanut butter product on the bread to evaluate spreadability and sample it to evaluate the other attributes.

Seventy-seven percent of panelists were females. The group was composed primarily of Caucasians (72%). Married panelists made up 62% of the sample. Most of the participants had completed high school and 58% had either attended college courses, completed college, or had an advanced degree. About 63% of the participants were employed full time, and the median annual household income was between $30,000 to $39,000 (Table 11).

Mean values for consumer scores for the control and HPNTB containing varying proportions of dry honey and salt were compared in Table 12. The control had a texture rating ($x = 6.8$) significantly higher ($p \leq 0.05$) than all HPNTB treatments. The treatment containing 20% dry honey and 3% salt had the lowest color rating ($x = 6.2$)

which was significantly different ($p \leq 0.05$) from the control. HPNTB containing 6% dry honey and 1% salt was not significantly different from the control except for texture ($x = 6.1$). Ratings for HPNTB containing 13% dry honey and 1% salt were significantly different from the control ($p \leq 0.05$) for spreadability, texture, and overall acceptability. Spreadability, texture, flavor, and overall acceptability scores of HPNTB containing 20% dry honey and 1% salt were significantly different than the control ($p \leq 0.05$).

Regression analysis results and the F-statistic were used to determine which reduced model was not significantly different from its corresponding full second order model. The best fitting models were used to generate contour plots (Fig. 5). The contour plots indicated that increasing salt percentage decreased the sweetness, flavor and saltiness ratings of the HPNTB. Spreadability, texture and overall acceptability rating decreased with increasing dry honey and salt percentage of HPNTB. The model for overall acceptance as a function of dry honey and salt (Overall acceptability - 7.0, 0.02 x_1 - 0.85 x_2, where x_1 = dry honey level and x_2 = salt level) showed that HPNTB containing 7% dry honey and 1% salt will have the rating of 6 (= like slightly).

Product formulations that were predicted to score 6.0 (like slightly) or higher for spreadability, flavor and overall acceptance were used to determine the optimum formulations. Contour plots were therefore superimposed. The areas of overlap (score = 6.0 or higher) were outlined (Fig 6). This area of overlap represents the region where all formulation would result in an acceptable product.

From the prediction equations, a HPNTB containing 7% dry honey and 1% salt would have the following scores: overall acceptability, 6.0; spreadability, 6.9; sweetness, 5.9; saltiness, 5.9; flavor, 6.1; and texture, 5.9. HPNTB containing a 7% dry honey and 1% salt would not be significantly different from the control for all attributes except the texture. Texture differences were found because the dry honey produced grainy mouthfeel.

Table 11. Selected demographic characteristics of participants in the consumer test on products containing peanut butter and honey (n = 93).

Item	Characteristic	Percent
Sex	Female	77.4
	Male	22.6
Age (years)	under 25	11.8
	25-34	26.9
	35-44	32.2
	45-54	18.3
	55-64	10.8
Race	White	72.0
	Others	28.0
Marital status	Single	15.0
	Married	62.4
	Others	22.6
Educational attainment	< high school	5.4
	completed high school	36.5
	some college	31.2
	college or higher	26.9
Employment status	employed full time	63.4
	employed part time	9.7
	unemployed	2.2
	others (homemaker retired, student)	24.7
Household income	under $20,000	22.6
	$20,000 - $29,999	16.1
	$30,000 - $39,999	23.7
	$40,000 - $49,999	17.2
	$50,000 - $59,999	9.7
	$60,000 and over	10.7

Table 12. Mean consumer scores for acceptability of honey peanut butter and a control (n = 93)

% by weight of ground peanut		Acceptability score[x]						
Honey	Salt	overall	color	spreadability	sweetness	saltiness	flavor	texture
6	1.0	6.05ab[y]	6.71a	6.96ab	5.98a	6.03ab	6.03ab	6.12b
6	1.5	5.59bc	6.58ab	6.80bc	5.53bc	5.54bc	5.54bc	5.73bc
6	2.0	5.17cd	6.65a	6.52bcd	5.18c	4.88d	5.14c	5.61bc
13	1.0	5.85b	6.74a	6.37cd	5.95ab	6.01ab	6.00ab	5.28cde
13	1.5	5.61bc	6.58ab	6.63bc	5.75b	5.59b	5.82b	5.66bc
13	2.0	5.05d	6.56ab	6.10de	5.04c	4.70d	5.13c	4.83ef
20	1.0	5.65bc	6.47ab	6.05de	5.89ab	5.83ab	5.70b	5.04def
20	1.5	5.68bc	6.59a	5.87ef	5.94ab	5.76b	5.87b	5.40cd
20	2.0	4.76d	6.19b	5.55f	5.10c	5.05cd	5.01c	4.65f
control[z]		6.43a	6.84a	7.34a	6.23a	6.25a	6.46a	6.77a

[x]A 9-point hedonic scale was used (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely).
[y]Mean values in the same column that are not followed by the same letter are significantly different (a = 0.05).
[z]control = regular peanut butter without honey contain 6% corn syrup solids and 1% salt.

2. Enzymatic and Autoxidation of Defatted Peanuts

Model systems which utilize pure reactants and conducted under controlled test conditions can give an indication of enzymatic and autoxidation reactions that take place in peanuts. It is, however, difficult to fully relate these reactions to full-fat and partially defatted peanuts subjected to environmental conditions to which they are exposed in the marketplace. It was the objective of this study to determine the effect of decreasing oil content in raw and roasted peanuts on the enzymic and autoxidation of the samples, respectively. Specifically, we investigated the oxidative stability of full-fat and defatted raw and roasted peanuts by established methods and the Oxidation Stability Index (OSI) method. The sensory lipid oxidation characteristics of defatted roasted peanuts were also determined. Models for predicting responses of the dependent variable (OSI) from the independent variable (% oil) in raw and roasted peanuts were developed.

Table 13 shows the standards and scores used as sensory descriptors. Roasted peanuts had lower peroxide values compared to raw peanuts. Roasted full-fat peanuts oxidized faster than raw peanuts. Raw peanuts stored at 4°C exhibited appreciable peroxidation only after the 5th month, but still proceeded more slowly than that of roasted peanuts.

Descriptive sensory results are shown in Fig. 7. Regression analysis revealed that only roasted peanutty, burnt odor, and peanutty, rancid/oxidized and burnt flavor descriptors differed significantly with oil content of peanuts. Models that predict the responses of dependent sensory variables from the independent variable (% oil) of roasted peanuts are presented in Table 14.

The peroxide values of ground, fresh, raw peanuts that had undergone enzymatic oxidation decreased significantly with decreasing oil content, indicating greater oxidative stability with decreasing oil content. The peroxide values of ground, rancid, roasted peanuts that had undergone autoxidation likewise significantly decreased with decreasing oil content. A high OSI value for raw peanuts indicated greater stability than roasted peanuts at all oil levels. Roasted peanutty and burnt odor, and roasted peanutty, rancid/oxidized and burnt flavor scores of ground, rancid, roasted peanuts decreased with decreasing oil content.

Table 13. Standards and scores used for the descriptors

Descriptors	Standard	Score
Odor		
Roasted peanutty	Planters roasted peanuts	7.0
Rancid/Oxidized	Rancid peanut oil	7.5
Cardboardy	Moist cardboard	3.0
Burnt	Burnt toast[a]	7.0
Flavor		
Roasted peanutty	Planter's roasted peanuts	7.0
Rancid/Oxidized	Rancid peanut oil	2.5
Cardboardy	Moist cardboard	8.5
Burnt	Burnt toast[a]	10.0

[a] 8 slices of loaf bread oven-heated at 288°C for 14 min.

Table 14. Models that predicted the responses of dependent sensory variables from the independent variable % oil of roasted peanuts

Response Variable	Model	R^2
Odor		
Roasted peanutty	6.55 - 0.181 (% oil) + 0.004 (%oil)	0.8829
Burnt	-0.995 + 0.053 (% oil)	0.6644
Flavor		
Roasted peanutty	12.806 - 0.601 (% oil) + 0.009 (% oil)^2	0.7886
Rancid/Oxidized	-4.34 + 0.400 (% oil) - 0.006 (% oil)^2	0.5017
Burnt	6.822 - 0.405 (% oil) + 0.006 (% oil)^2	0.8836

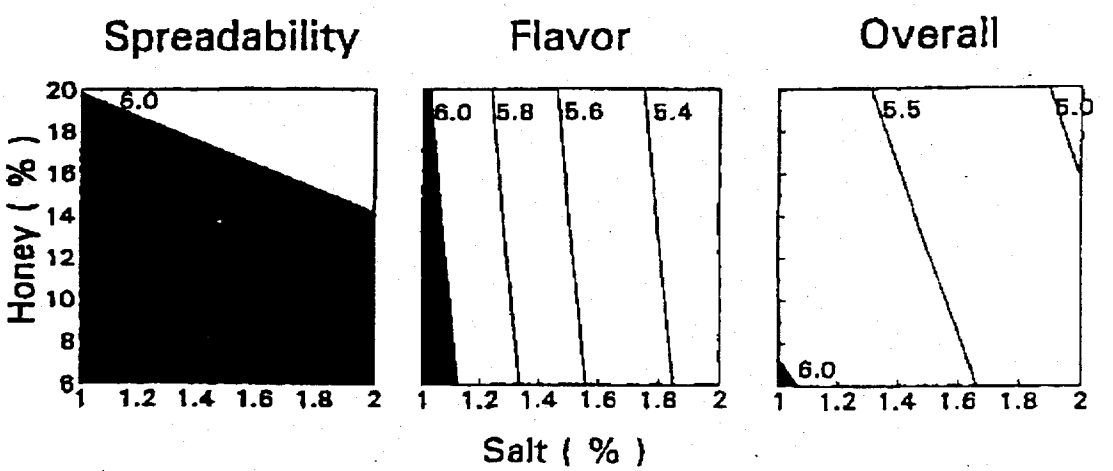

Figure 5. Contour plots for spreadability, flavor and overall acceptance of a honey-peanut butter spread.

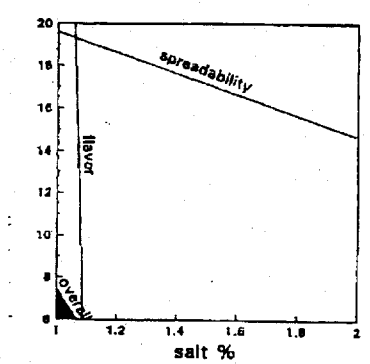

Figure 6. Superimposed contour plots for spreadability, flavor, and overall acceptance of a honey-peanut butter spread. The shaped region represents all formulations that would result in acceptable product.

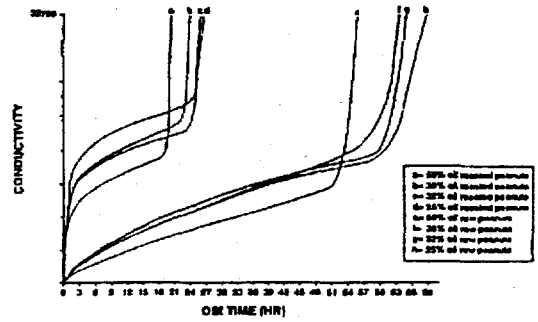

Figure 7. OSI time as affected by conductivity of roasted and raw peanuts.

3. Kinetic Modeling of Lipid Oxidation of Dry-Roasted Peanuts

Studies were conducted to determine suitable kinetic models for lipid oxidation in dry roasted peanuts. The implications of such a study are in the development of appropriate storage, handling, packaging or pretreatment techniques for storage quality maintenance of peanuts. Three models (Maloney, Labuza and Koelsch) were evaluated.

Maloney Model: To describe the kinetics for the initial oxidation stages when monomolecular decomposition of hydroperoxides predominates, the following rate equation was formulated by Maloney and colleagues in 1966.

$$\frac{-d[O_2]}{dt} = \frac{d[ROOH]}{dt} = \frac{k_p k_i^{1/2}}{(2K_t)^{1/2}}[M]^{1/2}\,[RH][ROOH]^{1/2}$$

Demonstrating that the concentration of the metal catalyst and the lipid substrate did not change significantly, the equation simplified to

$$[-O_2]^{1/2} = [ROOH]^{1/2} = 0.5\,K_m t$$

$$\text{where } K_m = \frac{k_p k_i^{1/2}}{(2k_t)^{1/2}}[M]^{1/2}\,[RH]$$

Labuza Model: Considering the overall reaction taking place, a different model was developed by Labuza in 1971.

$$\frac{-d[O_2]}{dt} = \frac{d[ROOH]}{dt} = \frac{R_i^{1/2}}{k_t^{1/2}}\,\frac{k_p[RH]\,k_o[O_2]}{(k_p[RH] + k_o[O_2])}$$

When the lipid content becomes negligible to that of oxygen, the model was simplified to a zero order reaction.

$$-[O_2] = [ROOH] = K_L t$$

$$\text{where } K_L = \frac{R_i^{1/2} k_p}{k_t^{1/2}}[RH]$$

Koelsch Model: On the assumption that the rate of hexanal production is directly proportional to the peroxide content, another model was derived.

$$[Hex] = [Hex]_o + \frac{K_k^3 t}{3}$$

where $K_k = K_h K_m$, and
K_h = rate of hexanal formation

Peanuts (Virginia type) were dry roasted at 163°C for 30 min in an electric roaster. The peanuts were then cooled, blanched, cleaned, divided into four batches and stored at 30, 40, 50 or 63°C. Samples were analyzed periodically for 16 weeks or until a hexanal level of 15 ppm for peroxide value, total lipids, conjugated diene hydroperoxides (CDHPs), hexanal content and thiobarbituric acid reactive substances (TBARS). According to reports in the literature, one day at 63°C is equivalent to 1 week at ambient conditions. Thus, 16 weeks at 63°C would approximately be equivalent to 2 years at ambient temperature. A hexanal content of 0.15 ppm is considered as a detectable threshold of lipid oxidation; however, this value does not necessarily correspond to unacceptable levels of rancidity. A 100-fold threshold level was the criterion for discontinuing the storage study.

Figs. 8 to 12 show changes in chemical parameters during storage. In all of these parameters, the interaction between temperature and time was statistically significant. The total lipid content (Fig. 8) was expected to be constant during storage. However, because of limitations of the chloroform:methanol extraction procedure, this was not observed. Changes in peroxide value (Fig. 9) did not follow a curve normally seen in oils. Since food systems were being studied, unlike in pure oil, the hydroperoxides did not accumulate, but instead were degraded as soon as they were formed. The TBARS (Fig. 10) did not show any consistent trend among different storage temperatures. The CDHP content (Fig. 11) increased rapidly as temperature increased. Previous studies revealed that the formation of CDHPs was proportionate to the uptake of oxygen. Thus, CDHP content was used as an index during the model fitting changes in hexanal content (Fig. 12) were similar to those of CDHPs, so hexanal was used as an index of hydroperoxide formation.

Table 15 summarizes the results of fitting the CDHP and hexanal data to the kinetic models. For the Maloney and Labuza models, R^2 values from the CDHP data were generally lower than those from the hexanal data, showing that the latter was a better indicator of lipid oxidation. All models especially the Maloney and Koelsch models, yielded higher R^2 values from this chemical parameter. Using various rate constants from the three models, the natural log of these constants was plotted against the reciprocal of storage temperature (in Kelvin), following the Arrhenius relationship. Table 16 shows the results of these correlations. All models had relatively high R^2 values, 0.87 to 0.91. From these models, the rate constant can be predicted at any given storage temperature, and vice versa.

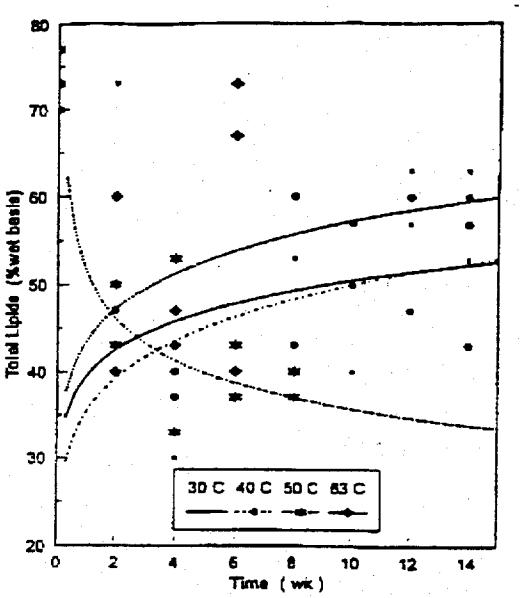

Figure 8. Changes in total lipid content during storage

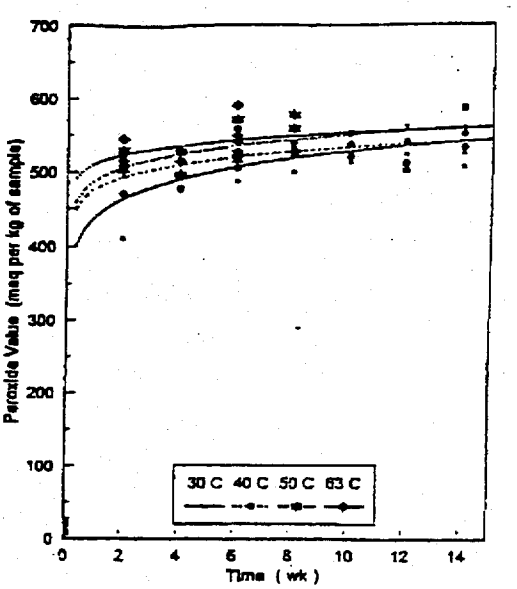

Figure 9. Changes in peroxide value during storage

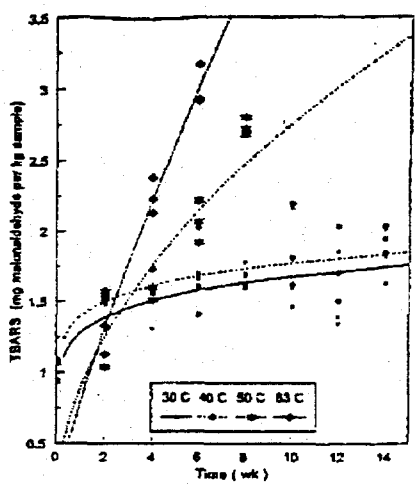

Figure 10. Changes in thiobarbituric acid reactive substance (TBARS) content during storage.

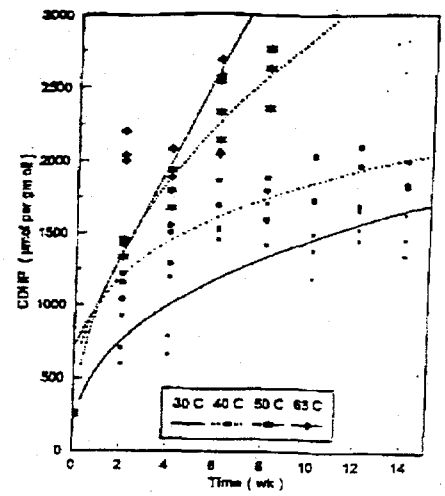

Figure 11. Changes in conjugated diene hydroperoxide (CDHP) content during storage.

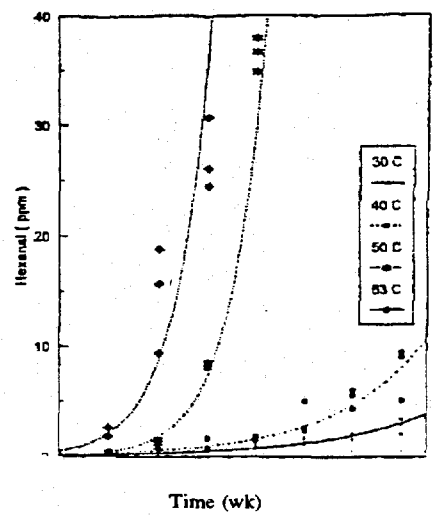

Figure 12. Changes in hexanal content during storage

4. Formation of Peanut Protein Films

Experiments were conducted to investigate formulation characteristics of peanut films using two methods, surface film formation and deposition techniques. Materials used in this study are listed in Table 17. Raw peanuts (Virginia type) were roasted for 14 min at 163 °C in an electric roaster (model 37, Preedit Electric Roaster Co., Erie, PA). Skin and testae were removed by a roller type blancher and testae were separated from the kernels by air aspiration. Kernels were visually inspected and sorted, then kept in plastic bags at 4 °C until used. Defatted peanut flours were prepared by hexane extraction. Peanut protein concentrates were prepared from defatted peanut flour. Moisture, protein and fat content of raw peanut and flour were also analyzed (Table 18) to determine protein and lipid ratio. Procedures for preparing films are Figs. 13. and 14. Surface film formation was studied using various forms of starting material (Table 15). The deposition method was examined only for protein concentrate, since preliminary studies showed that films could not be formed from other forms of peanut products.

a. Surface film formation.

Films from full-fat or 28% fat peanut flour formed 20-30 min. The first harvest was difficult to remove because films were weak due to their high content. After the first harvest, films were harvested every 20-30 min, depending upon the desired thickness.

Films were formed from 10% fat flour in 20-30 min; a large piece of film could be removed intact from the surface. This film looked less oily compared to film made from full fat and 28% fat material. From protein concentrates, film formed within 30 min; however, it was not possible to harvest the film successfully until after 1 or 2 hours. Film from protein concentrate was more flexible and clear than that made from peanut flour. The solution with a high protein:lipid ratio was suitable for making film by this method. The color of these films varied from yellowish white to pale yellow, depending on the color of raw material. Also, it was possible to obtain films large enough for conducting physical and permeability tests, which was not the case for full fat, partially defatted or defatted raw materials.

b. Deposition method.

Films could be formed when the pH of the solutions ranged from 6 to 11. At pH 5, film was not formed, probably because this is close to the isoelectric point of peanut protein. All films had a brownish yellow color, but became darker and clearer as the pH increased. Film could be formed without a plasticizer but was very brittle. At least 5 to 10 g of plasticizer per 100 g of solution was needed. Preheating the solution before film casting resulted in clearer films. The effect of various casting materials was also investigated. Films could be peeled off of glass and plastic dishes but they stretched during the peeling process. When an aluminum foil laminated glass dish was used, it was easier to peel the film off the aluminum foil than from the glass dish directly. The smoothness of the film surface depended on the casting surface. A Teflon coated pan worked very well except that bubbles formed on the bottom side of the film. It is believed that this occurred due to the high rate of film formation and high thermal conductivity of the Teflon coated pan. Oven (70°C), vacuum (50°C) and ambient temperature were studied for drying the films. Film could be formed when the oven and vacuum drying methods were employed but not at ambient temperature. A high temperature was needed in the deposition method to facilitate protein denaturation. Film subjected to vacuum drying contained a lot of bubbles and the surface of the film was not smooth.

Films made using the deposition method were more flexible than those made using the surface formation method these films were clear to lower carbohydrate content. The color of these films varied from light to dark brownish yellow depending on the color of the concentrate.

Table 15. Rate constants for various lipid oxidation models using CDHP and hexanal data

Temp. (°C)	Rate Constant for Three Models*				
	Maloney		Labuza		Koelsch
	CDHP (K'_m)	Hexanal (K''_m)	CDHP (K'_L)	Hexanal (K''_L)	Hexanal (K_k)
30	2.87 (0.66)	0.23 (0.96)	83.98 (0.67)	0.19 (0.86)	0.0022 (0.93)
40	3.04 (0.60)	0.38 (0.92)	97.76 (0.66)	0.53 (0.77)	0.0062 (0.93)
50	8.05 (0.84)	1.44 (0.87)	279.95 (0.91)	4.05 (0.67)	0.1895 (0.95)
63	9.79 (0.68)	1.79 (0.97)	318.90 (0.70)	4.69 (0.89)	0.3915 (0.96)

* Values in parenthesis refer to R^2 (coefficient of determination) for the model

Table 16. Arrhenius constants of kinetic models

Kinetic Model	Data Used	R^2 *	Arrhenius Constant
Maloney	CDHP	0.87	4312
Maloney	Hexanal	0.91	6981
Labuza	CDHP	0.87	4671
Labuza	Hexanal	0.89	10731
Koelsch	Hexanal	0.90	17575

* Coefficient of Determination

Table 17. Materials for preparing peanut protein films

Stock for film preparation		Film preparation method	Raw material and (source)
Code	Material		
RP	Roasted Peanut	Surface formation	Raw peanut (Jardina)*
PDF-28-G	Partially Defatted flour	Surface formation	28% fat flour (Golden)
PDF-28-C	Partially Defatted flour	Surface formation	Raw peanut (McCleskey)*
PDF-12-G	Partially Defatted flour	Surface formation	12% fat flour (Golden)
PDF-10-S	Partially Defatted flour	Surface formation	10% fat flour (Seabrook)
DF-G	Defatted flour	Surface formation	12% fat flour (Golden)*
DF-S	Defatted flour	Surface formation	10% fat flour (Seabrook)*
PC-G	Protein concentrate	Deposition method	12% fat flour (Golden)*
PC-S	Protein concentrate	Deposition method	10% fat flour (Seabrook)*

* Further processing done at CFSQE

Table 18. Proximate composition of raw materials

Material	Moisture (wb)	Protein (db)	Fat (db)	Protein/lipid
RP	4.32	26.43	46.97	0.56
PDR-28-G	3.50	46.63	29.02	1.61
PDR-12-G	3.50	57.62	12.58	4.58
DF-G	1.94	64.88	1.39	46.68
PC-G	1.98	76.18	0.26	293.00
PDF-28-C	7.67	38.38	28.01	1.37
PDF-10-S	2.44	47.86	10.31	4.64
DF-S	2.04	50.35	1.94	25.95
PC-S	4.16	84.90	1.13	75.13

5. Development of a Low Fat Beverage from Peanuts

Two separate studies are reported here as a part of the ongoing process development for a peanut-based low-fat beverage. To prepare beverages, partially-defatted roasted peanuts were milled and wet-blended, slurries were filtered through screens with 34 μm mesh size, and filtrates were blended with sugar, salt and emulsifier (Fig. 15). Beverages were homogenized, bottled and then heat processed (bottle-processed at 72°C for 2 min or 111°C for 8 min) or pasteurized in a kettle, homogenized and then bottled (kettle-pasteurized for 2 min at 72, 77 or 82°C).

a. Effect of heat processing protocols on selected physico-chemical properties of the beverage.

The objectives of this study were to evaluate the effects of two heat processing protocols on total solids, suspension stability, viscosity and color of a beverage prepared from partially-defatted roasted peanuts. Effects of heat processing on selected sensory attributes of the beverage were also studied. Presented in Table 19 are the physical prepared by the two

processing methods. Heat processing did not adversely affect the total solids (%) and color of the peanut beverage. Samples that were bottle-processed at 111°C were the most stable and 2-3 times more viscous than the kettle-pasteurized treatments. Trends observed for suspension stability were different from those reported for beverages prepared from unroasted peanuts and soybeans by other researchers. Suspension instability of our beverages may have been promoted by the defatting and filtering operations, and in the case of the kettle-pasteurized samples, by the high homogenizing temperatures used. These procedures decreased quantities of fat globules available for complexing, may have excessively denatured protein molecules, and possibly dissociated protein agglomerates and protein-lipid complexes that were formed during pasteurization. Preliminary sensory evaluation indicated that the kettle-pasteurized samples had stronger roasted peanut flavor and smoother mouthfeel than samples which were bottle-processed at 111°C. Kettle-pasteurization also resulted in products with viscosities which were within the range observed for commercial milks. Kettle-pasteurization using processing temperatures of 72-100°C with heating intervals longer than 2 min and homogenizing temperatures < 82°C shows potential for producing peanut beverages with physical and sensory attributes similar to commercial animal milks.

b. Effect of heat processing on particle size distribution of the beverage.

Knowledge of particle size in the heat-processed products is necessary to select processing parameters which would give a smooth beverage. The objectives of this study were to evaluate the effects of the two heat processing methods on particle size distribution in a beverage prepared from partially-defatted roasted peanuts. Fractional filtration was used to measure particle size in the peanut beverages. Particles (g/100 g total solids) with diameters

< 34 µm ranged from 98.4 to 99.4 in bottle-processed and from 98.8 to 99.3 in kettle-pasteurized samples (Table 20). Beverages which were bottle-processed at 111°C contained more particles with diameters of 104 to 124 µm than other peanut beverage samples or commercial low-fat and whole milks. Differences in particle size due to emulsifier type were observed only in samples which were bottle-processed at 111°C or kettle-pasteurized at 72°C. Distribution of particles retained on 53, 74 and 104 µm screens indicates that peanut beverages which were bottle-processed at 72°C or kettle-pasteurized at 72, 77 or 82°C would be less chalky than commercial chocolate low-fat milk and chocolate drink. Relative viscosities and particle retention on 34 µm screens suggest that peanut beverages kettle-pasteurized at 77 or 82°C would have optimum mouthfeel and may be similar in smoothness to commercial low-fat and full-fat cow's milks. Results indicate that beverages with little or no chalky mouthfeel can be produced when partially-defatted roasted peanuts are milled and wet-blended, and slurries are filtered through a 34 µ-m screen before formulations are kettle-pasteurized at 77 to 82°C and homogenized at 77 to 82°C.

Table 19. Physical properties* of peanut beverage after 7-day storage at 1°C.

			Bottle-processed[a]		Kettle-pasteurized[a]		
Property	Emulsifier	Control	72°C	111°C	72°C	77°C	82°C
Total Solids (%)	CM	12.6±0.02d	12.6±0.07d	12.6±0.01d	12.8±0.49cd	13.4±0.17b	13.3±0.41b
	HV	13.0±0.03c	13.0±0.03c	13.0±0.02c	13.9±0.27a	13.8±0.03a	13.8±0.28a
Viscosity (cps)	CM	2.4±0.06h	4.3±0.0g	17.4±0.0a	6.1±0.31f	6.4±0.91ef	6.5±0.54de
	HV	2.0±0.06j	2.1±0.0i	17.4±0.0a	8.4±0.44b	7.4±0.35c	6.7±0.06d
Emulsion stability index	CM	0.4±0.03de	0.4±0.04de	0.7±0.02a	0.4±0.04cde	0.5±0.07b	0.4±0.06bcd
	HV	0.4±0.03de	0.4±0.04cde	0.7±0.06a	0.5±0.04bc	0.5±0.05bc	0.4±0.05bc
Visual stability index	CM	0.6±0.01f	0.6±0.01f	0.9±0.01a	0.6±0.02cd	0.6±0.02e	0.6±0.02cde
	HV	0.5±0.00h	0.5±0.01g	0.8±0.01b	0.6±0.02c	0.6±0.02de	0.6±0.03cde
Hue Angle	CM	68.9±0.05cd	66.5±0.02fg	68.0±0.08de	70.8±0.00ab	69.9±0.09bc	69.4±0.02c
	HV	67.2±0.03ef	59.6±0.02i	64.7±0.08h	71.8±0.05a	71.2±0.03a	69.8±0.07bc
L value	CM	71.5±0.09a	65.2±1.37cde	72.2±0.15a	65.5±1.14cde	64.2±2.15def	63.9±1.04ef
	HV	69.1±0.09b	53.3±0.28g	65.4±0.48cde	65.4±1.63cde	66.3±1.91c	65.5±0.42cde
Chroma	CM	7.5±0.05de	7.2±0.03e	8.8±0.08ab	8.8±0.09ab	8.3±0.02abc	8.2±0.08bc
	HV	7.0±0.02e	4.7±0.01f	7.0±0.07e	9.0±0.06a	8.8±0.0ab	8.3±0.01abc

*Means ± standard deviation. Means within each parameter with the same letter are not significantly different at p=0.05.
+Emulsifiers: CM=carrageenan, HV=hydrogenated mono-diglyceride.
[a]Processing time at 111°C was 8 min. All other samples were processed for 2 min.

Table 20. Total solids* (g/100 mL) and solids* (g/100 g total solids) < 34 µm diameter in peanut beverage samples after 7-d storage at 1°C.

		Control	Bottle-processed[a]		Kettle-pasteurized[a]		
Property	Emulsifier+		72°C	111°C	72°C	77°C	82°C
Total Solids	CM	12.6±0.02d	12.6±0.07d	12.6±.01cd	12.8±0.49cd	13.4±0.17b	13.3±0.41b
(g/100 mL)	HV	13.0±0.03c	13.0±0.03c	13.0±0.02c	13.9±0.27a	13.8±0.03a	13.8±0.28a
Solids (g/100g	CM	99.3±0.05a	99.3±0.05a	99.1±0.05a	98.8±0.24b	99.2±0.04a	99.2±0.15a
total solids), <34 µm dia	HV	99.4±0.00a	99.4±0.05a	98.4±0.50c	99.3±0.06a	99.3±0.00a	99.3±0.01a

*Means (± standard deviations) of triplicate determinations. Means for the same property with the same letter are not significantly different at p≤0.05.
+Emulsifier type: CM=carrageenan, HV=hydrogenated mono-diglyceride
[a]Processing time at 111°C was 8 min. All other samples were processed for 2 min.

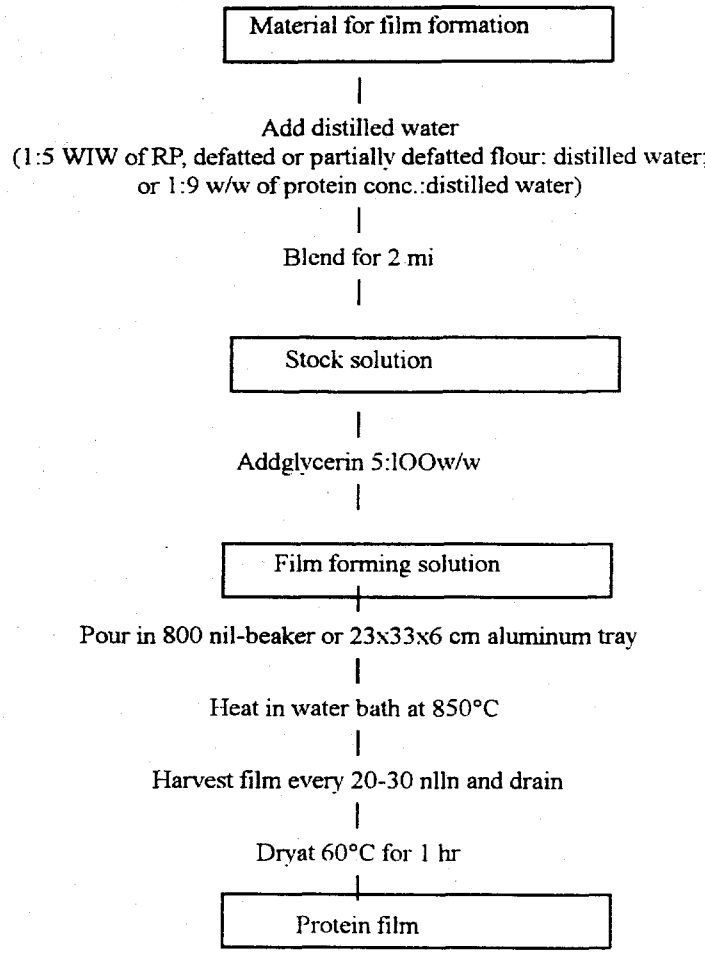

Figure 13. Flow chart for surface film formation

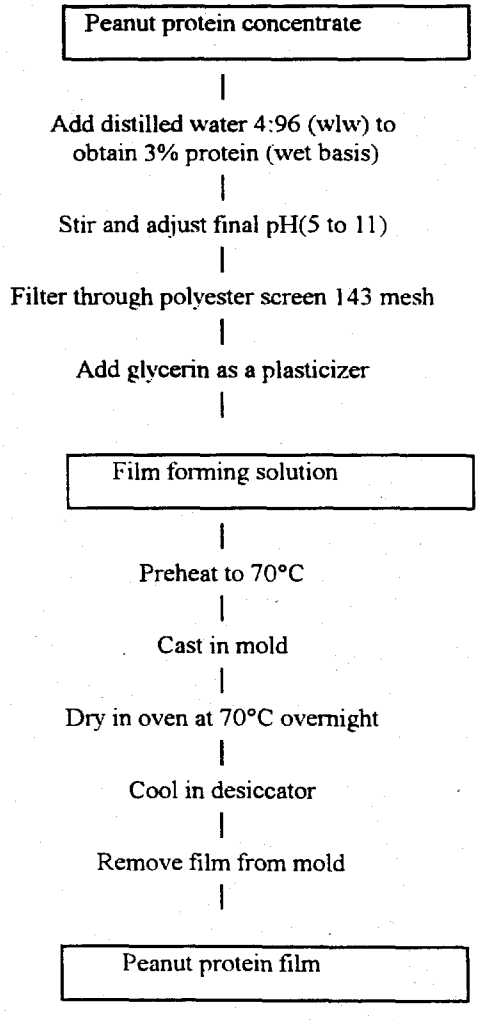

Figure 14. Flow chart for deposition method

6. Bacterial Degradation of Aflatoxin

Two sets of experiments were done in an effort identify the biochemical mechanism by which *Flavobacterium aurantiacum* degrades aflatoxin B_1 (AFB1). The first set of experiments was conducted to compare the ability of *F. aurantiacum* to degrade aflatoxin in the presence and absence of EDTA. Maximum degradation of AFB1 was obtained within 48 hr with 1×10^{13} cfu/ml (corresponding to approx. 2.5 mg/ml protein). When 2 mM EDTA was incorporated into phosphate buffer (pH 7.0) containing AFB1 at a concentration of 3 μg/ml, more degradation was observed than when cells were incubated with AFB1 alone. It is therefore assumed that EDTA has some effect on the metabolism/degradation of AFB1 by *F. aurantiacum*. It is probable that EDTA acts as a chelator and binds inhibitor (possibly divalent cations), interacts with AFB1 in some unknown manner or enhances cell metabolism to degrade AFB1 faster.

In the second set of experiments, the cellular location of the enzyme system(s) involved in the degradation of AFB1 was investigated. *F. aurantiacum* cells were lysed and separated into cytosolic and membrane fractions,

and the relative degradation of aflatoxin B_1 in each fraction was determined in each using a U.V. spectrophotometric method. Conclusive results were not obtained due to the presence of a high level background absorption by cytosolic and membrane fractions. Hence, this series of experiments will be repeated using HPLC for quantification of aflatoxin.

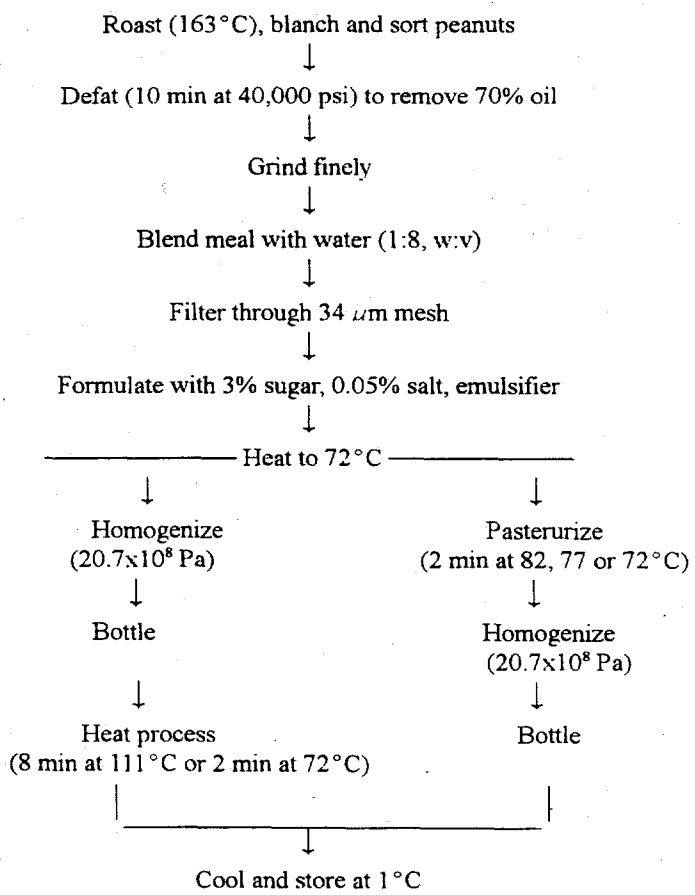

Figure 15. Flow diagram for preparation of peanut beverage

TRAINING ACCOMPLISHMENTS

In the area of training, Mr. Panuwat Suppakul, Ms. Wimolsiri Thanavisuthra and Ms. Supang Reungchay are making good progress toward meeting the requirements of their M.S. degrees at the Department of Product, Development, Kasetsart University.

As part of the technology transfer activities of the project, at Kasetsaart University, Peanut CRSP researchers cooperated with home economists from the Department of Agriculture Extension, Ministry of Agriculture to transfer technology of ground roasted peanut, hot sand roasted peanut and coated peanut products to housewives at Kud-Jub District, Udorn-thanee Province. Housewives were very receptive and are producing these products for markets in their communities.

A workshop on Training the Trainers from 13 countries on the topic of "Quality Evaluation and Utilization of Food Legumes" was held at Kasetsart University from April 24 to May 27, 1995. The Workshop was attended by 20 participants, from 13 countries: Tanzania (2), Fiji Island (1), Srilanka (1), Republic of Maldives (1), Bhutan (2), Bangladesh (2), Sierra Leone (2), Loas P.D.R.(2), Cambodia (1), Korea (1), Vietnam (2), Nepal (1) and Thailand (2). All participants were government officers whose work is related to legumes utilization. Proceedings were distributed at the Workshop. Many examples given during the training program were peanut products developed by the Peanut CRSP project. Cooperation with the Department of Technical and Economic Cooperation, Office of Prime Minister under the Thai-AID project facilitated the training course. This training course will be conducted annually and will result in a large impact on processing of legumes as a whole and the nutritional well-being of consumers throughout Southeast Asia as well as in other developing countries in the world in future years.

Cooperators at the Guadalupe Women's Association and URIVAC were trained in processing, management and marketing of peanut-extended snacks be Peanut CRSP investigators at ViSCA. Sessions were held at ViSCA as well as in rural areas in the Baybay vicinity.

Dr. Anna Resurreccion trained staff members of the Food Development Center (FDC) of the National Food Authority, Ministry of Agriculture In Taguig, Metro-Manila, Philippines on sensory evaluation methodology, specifically on descriptive analysis techniques and consumer affective testing procedures.

Training courses on sensory evaluation and quality control and consumer testing of food products were conducted by Dr. Resurreccion on November 28 to December 2, 1994 and in the Malaysian Agricultural Research and Development Institute (MARDI) in Serdang, Selangor, Malaysia. A total of 105 (35 in the Philippines and 60 in Malaysia) participants, mostly from the food industry and a few from government agencies and academic institutions, enrolled in the courses. The courses were sponsored by USAID (PITO Project), Technonet Asia and the FDC and MARDI in the Philippines and Malaysia, respectively.

Mr. Witoon Prinyawiwatkul (Thai), Mr. Anuvat Jangchud (Thai) and Ms. Doris D'Souza are pursuing Ph.D. degree programs at the University of Georgia.

Mr. Agustin Ramos (Filipino) is pursuing an M.S. degree at the University of Georgia. Ms. Grace Divino (Filipino) completed her M.S. degree and entered the Ph.D. program at Kansas State University.

Dr. Margaret J. Hinds left her position as a Post Doctoral Associate at the University of Georgia to a take position as Assistant Professor at North Carolina A&T University. Dr. Hinds' support was from two Peanut CRSP projects (GA/PH/CAR and GA/FT/TP).

PRESENTATIONS AND CONFERENCE PARTICIPATION

Beuchat, L. R. 1994. Advances in food mycology methodology. (Abstr.) East, Central and Southern Africa Food Science and Technology Conference '94: Challenges for Africa Towards the Year 2000. September 12-16, Victoria Falls, Zimbabwe.

Beuchat, L. R. 1994. Detection and enumeration of microorganisms in hurdle technology foods, with particular consideration of foods with reduced water activity. Int. Symp. Properties of Water. Practicum II, Food Preserv. by Moisture Control. 19-24 June, Puebla, Mexico.

Chinnan, M. S. 1995. Improving peanut postharvest systems. Impacts and Scientific Advances through Collaborative Research on Peanut. International Research Smyposium and Workshop of the Peanut Collaborative Research and Support Program. Washington, D.C., March 29-31.

Chompreeda, P., V. Haruthaitanasan and W. Chantrapornchai. 1995. Development of a vegetarian product from texturized peanut protein. Ann. Mtg. IFT June 3-7, Anaheim, CA.

Garcia, V. V., R. L. Natividad, M.G.L. Bailon and C. DV. Almario. 1994. Development of Soft Curd from Peanuts. Poster paper presented at IFT Annual Meeting and Food Expo. Atlanta, Georgia, USA. June 25-29.

Garcia, V. V. and L. R. Beuchat. 1995. Monitoring Network to Survey Food Products for Aflatoxin in the Philippines. Workshop on Impacts and Scientific Advances through Collaborative Research on Peanut, Peanut-CRSP Internationnal Research Symposium and Workshop, Arlington, VA.

Haruthaithanasan, V. and P. Chompreeda. 1995. Technology Transfer manual on Utilization of Peanut to housewives at Kud-Jub District, Udorn Thanee Province, 12-17 March, Dept. Of Product Development, Faculty of Agro-Industry, Kasetsart University, Bangkok.

Haruthaithanasan, V. and P. Chompreeda. 1995. Women Cooperative in Peanut Processing in Thailand. Paper presented at the Peanut Workshop, Washington, D.C., U.S.A., 29-31 March, Department of Product Development, Faculty of Agro-Industry, Kasetsart University, Bangkok.

Hashim, I. and A.V.A. Resurreccion. 1995. Optimization of honey-peanut butter for consumer acceptance. Ann. IFT Mtg., Anaheim, CA, June 3-7.

Hinds, H. J. 1995. Quality enhancement and technology transfer of peanut butter. Impacts and Scientific Advances through Collaborative Research on Peanut. International Research Symposium and Workshop of the Peanut Collaborative Research Support Program. Washington, D.C., March 29-31.

Hinds, H. J., L. R. Beuchat and M. S. Chinnan. 1994. Particle size distribution in a peanut-based beverage. (Abstr.) American Peanut Research and Education Society Meeting, Tulsa, OK, July 12-14.

Hinds, M. J., M. S. Chinnan, and L. R. Beuchat. 1995. Acceptance and purchase intent of peanut butter by Belizean consumers. (Abstr.) Ann. Mtg., Institute of Food Technologists, Anaheim, CA, June 3-7.

Hinds, M. J., L. R. Beuchat and M. S. Chinnan. 1994. Particle size distribution in a peanut-based beverage. (Abstr.) Annu. Mtg. Amer. Peanut Res. Educ. Soc., July 13-15, Huntsville, AL.

Makesa, R. C. 1995. Microbiological control of aflatoxigenic fungi and other organisms in foods. Manila Commission Diamond Jubilee Professional Charr lecture, June 21, Manila.

Resurreccion, A.V.A. 1994. Sensory Evaluation in Quality Control--Background information and initial program steps. Workshop on Sensory Evaluation in Quality Control and Consumer Testing. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines, November 28.

Resurreccion, A.V.A. 1994. Implementation of a quality control/sensory evaluation program--Comprehensive Descriptive Method. Workshop on Sensory Evaluation in Quality Control and Consumer Testing. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines, November 29.

Resurreccion, A.V.A. 1994. Consumer sensory evaluation/affective tests. Workshop on Sensory Evaluation in Quality Control and Consumer Testing. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines, November 30.

Resurreccion, A.V.A. 1994. Implementation of a quality control/sensory evaluation program--Quality Ratings Method Workshop on Sensory Evaluation in Quality Control and Consumer Testing. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines. December 1.

Resurreccion, A.V.A. 1994. Implementation of a quality control/sensory evaluation program--Degree of Difference from Control Method, Workshop on Sensory Evaluation in Quality Control and Consumer Testing. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines. December 1.

Resurreccion, A.V.A. 1994. Implementation of a quality control/sensory evaluation program--In/Out Method. Workshop on Sensory Evaluation in Quality Control and Consumer Testing.. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines. December 2.

Resurreccion, A.V.A. 1994. Data Analysis and Reporting of Sensory Evaluation Data. Workshop on Sensory Evaluation in Quality Control and Consumer Testing. Food Development Center, National Food Authority, Department of Agriculture, Manila, Philippines. December 2.

Resurreccion, A.V.A. 1995. Quantification of quality attributes as perceived by the consumer. Food Science and Human Nutrition Section, Southern Association of Agricultural Scientists Annual Meeting, New Orleans, LA, January 31.

Resurreccion, A.V.A. 1995. Sensory evaluation for peanut product development. Symposium on Impacts and Scientific Advances Through Collaborative Research On Peanut. U.S. Agency for International Development, Peanut Collaborative Research Support Program. Washington, D.C., March 29-31.

Wattanapat, R., T. Nakayama, L. R. Beuchat and R. D. Phillips. 1994. Kinetics of defatted peanut flour hydrolysis. (Abstr.) 5th ASEAN Food Conference, Food Agenda 21st Century, Kuala Lumpur, Malaysia, July 26-29.

INTERNATIONAL TRAVEL

Drs. Vichai Haruthaithanasan (Thailand), Virgilio Garcia (UPLB) and Luth Palomar (ViSCA) traveled to the U.S. (29 March-7 April) to attend a Peanut Workshop in Washington D.C., where they presented a paper reporting project activities.

Dr. Penkwan Chompreeda traveled to U.S. (June 1-8) to attend an Institute of Food Technologists 1995 workshop on "Optimizing Food and Beverage Flavors" and the 1995 Annual Meeting of IFT in Anaheim, California, where she presented a paper reporting project activities. At the IFT meeting, her paper entitled "Development of a Vegetarian Product from Texturized Peanut Protein" received the first place award in the International Research Paper Competition.

Dr. Anna Resurreccion traveled to the Philippines to hold workshops in the Philippines and Malaysia (funded by USAID, PITO Project and Technonet Asia) and to discussed research progress with Dr. Virgilio Garcia at the University of the Philippines at Los Baños.

PUBLICATIONS

Beuchat, L. R. 1995. Application of biotechnology to indigenous fermented foods. Food Technol. 49:97-99.

Beuchat, L. R. 1995. Advances in food mycology. Proc. East, Central and Southern Africa Food Science and Technology Conference '94:Challenges for Africa Towards the Year 2000, Victoria Falls, Zimbabwe.

Chompreeda, P., L. R. Beuchat, V. Haruthaithanasan and W. Chantrapornchai. 1995. Development of a vegetarian product from texturized peanut protein. (abstr.) Ann. Mtg. IFT June 3-7, Anaheim, CA. p. 88.

Clavero, M.R.S., Y.-C. Hung, L. R. Beuchat and T. Nakayama. 1994. Flavor, color and texture of peanuts treated with hydrogen peroxide. Peanut Sci. 21:1-4.

Divina, R. D. 1995. Isolation and Characterization of the Antagonistic Compounds from *Cladosporium fulvum* against toxigenic fungi. Ph.D. Dissertation, UP Los Baños.

Faculty, USAID-Peanut CRSP, Department of Product development. 1995. International Training Course on Quality Evaluation and Utilization of Food Legumes. 24 April-27 May, Dept. of Product Devel., Fac. of Agro-Industry, Kasetsart Univ., Bangkok. 204 pp.

Haruthaithanasan, V. and P. Chompreeda. 1995. Women Cooperative in Peanut Processing in Thailand. Paper presented at the Peanut Workshop, Washington D.C., U.S.A., 29-31 March , Department of Product Development, Faculty of Agro-Industry, Kasetsart University, Bangkok. 13 pp.

Haruthaithanasan, V. and P. Chompreeda. 1995. Technology Transfer manual on Utilization of Peanut to housewives at Kud-Jub District, Udorn Thanee Province, 12-17 March, Dept. of Product Development, Faculty of Agro-Industry, Kasetsart University, Bangkok. 20 pp. (In Thai).

Hinds, M. J., L. R. Beuchat and M. S. Chinnan. Physical properities of a heat-processed beverage prepared from roasted peanuts. (In press)

Hinds, M. J., M. S. Chinnan and L. R. Beuchat. Particle size distribution in a heat-processed beverage prepared from roasted peanuts. Lebensm. Wiss. Technol. (In press)

Hinds, M. J., M. S. Chinnan and L. R. Beuchat. 1994. Unhydrogenated palm oil as a stabilizer for peanut butter. J. Food Sci. 59:816-820, 832.

Line, J. E., R. E. Brackett and R. E. Wilkinson. 1994. Evidence for degradation of aflatoxin B1 by *Flavobacterium aurantiacum*. J. Food Prot. 57:788-791.

Line, J. E. and R. E. Brackett. 1995. Factors affecting aflatoxin B1 removal by *Flavobacterium aurantiacum*. J. Food Prot. 58:91-94.

McWatters, K. H., A.V.A. Resurreccion, L. R. Beuchat and R. D. Phillips. 1995. Use of peanut and cowpea in wheat-based products containing composite flours. Plant Foods Human Nutr. 47:71-87.

Mirabolo, J. 1994. Product Development of Peanut Sauce. B.S. Food Technology Thesis. UP Los Baños.

Palomar, L. S., A.V.A. Resurreccion, F.C.F. Galvez and L. R. Beuchat. 1994. Optimization of a peanut-sweet potato cookie formulation. Lebensm. Wiss. Technol. 27:314-318.

Prinyawiwatkul, W., L. R. Beuchat, R. D. Phillips and A.V.A. Resurreccion. Effects of peanut flour, feed moisture content and extrusion temperature on physical properties of an extruded snack product. Int. J. Food Sci. Technol. (In press)

Prinyawiwatkul, W., K. H. McWatters, L. R. Beuchat and R. D. Phillips. 1994. Physical properties of cowpea paste and akara as affected by supplementation with peanut flour. J. Agric. Food Chem. 42:1750-1756.

Wattanapat, R., T. Nakayama and L. R. Beuchat. 1995. Characteristics of acid hydrolysate made from defatted peanut flour. J. Food Sci. 60:443-445.

Wattanapat, R., T. Nakayama, L. R. Beuchat and R. D. Phillips. 1994. Kinetics of acid hydrolysis of defatted peanut flour. J. Food Sci. 59:621-625.

Wattanapat, R., T. Nakayama, L. R. Beuchat and R. D. Phillips. 1994. Kinetics of defatted peanut flour hydrolysis. (Abstr.) 5th ASEAN Food Conference, Food Agenda 21st Century, Kuala Lumpur, Malaysia, July 26-29. P. 306-307.

PLANS FOR 1995-96:

A. Kasetsart University

1. Continue to develop a breakfast bar using undersized peanut as a source of protein. Shelf-life and consumer acceptance of this product will be evaluated.

2. Continue to implement the utilization of peanut tempeh in food by housewives at village level.

3. Continue to utilize peanut hulls in development of a mosquito repelling stick.

4. Cooperate with the Department of Agriculture Extension, Ministry of Agriculture to transfer technology of peanut processing to home economists and industrials in Northern and Northeastern areas.

5. Cooperate with the Department of Technical and Economic Cooperation, Office of Prime Minister to offer a training course on quality evaluation and utilization of legumes to train participants from Indo-China countries and other countries under Thai-AID projects. The training period will be 5-week for 20 participants from those countries.

B. University of The Philippines at Los Baños

1. Characterization of the compound(s) produced *Cladosporium fulvum* that inhibit the growth of aflatoxigenic aspergilli will continue.

2. Technical assistance to small-scale peanut handling and processing industries for the purpose of improving quality and safety (aflatoxin contamination) of peanut products will continue.

C. Visayas State College of Agriculture

1. Work on peanut-based snacks and confectionery products will continue.

2. Equipment and tools for small-scale drying of peanuts and for processing peanut-based products at women's cooperatives will be developed and evaluated.

3. Technologies to increase utilization of peanuts in value-added products will be transferred to small- and medium-sized cooperatives operated and managed by women.

D. University of Georgia

1. Work will be done to develop an acceptable model for characterizing flavor changes in full-fat and defatted roasted peanuts during storage by correlating 4-6 volatile compounds with subjective sensory scores.

2. Research will be initiated on the flavor enhancing properties and nutritional contribution of peanut meal/flour and fermented meal/flour in food systems.

3. The suitability of using edible films on lipid oxidation of roasted, stored peanuts will be investigated.

4. Physical, chemical and sensory properties of a beverage prepared from roasted peanuts using various processing technologies will be determined.

5. Snack products containing flours prepared from partially defatted peanuts and cowpeas subjected to solid-state fungal fermentation will be formulated and characterized for texture, color and flavor qualities.

6. Physical and mechanical properties of selected peanut cultivars as related to handling, storage and processing will be studied.

7. An interactive computer model to design peanut drying systems will be developed.

PROGRESS TOWARD RESOLVING CONSTRAINTS

Progress in relieving constraints associated with peanut utilization and consumption has been made in the areas of socioeconomics, health and nutrition and research capacity.

1. Socio economic

Surveys of consumers in The Philippines have revealed the position peanut plays in the economy and the diet. The impact of peanut on the nutritional status of consumers as well as the economic well-being of small processors of peanut products has been more clearly defined.

Transfer of peanut processing technologies to rural areas of The Philippines and Thailand has enhanced the economic status of these groups. Consumer surveys are being used to critically assess the role of peanut in the diets of various income-level groups. Stategies to enhance the nutritional quality of diets have been developed and are being introduced to the consumer.

Significant progress has been made in reducing the amount of aflatoxin in peanut and peanut product made available to the consumer in Thailand and The Philippines. This has been accomplished by instructing peanut handlers and processors in procedures for reducing the potential of aflatoxin formation during storage and reducing the risk of the presence of aflatoxin in processed peanut products. Through a continuing series of articles in a daily Bangkok newspaper and through extension activities directed toward peanut farmers, handlers, and processors in The Philippines, tremendous progress has been made in making Thais and Filipinos aware of health implications associated with aflatoxin. A federal regulatory agency in The Philippines has, in fact, demanded the removal of a aflatoxin-contaminated peanut butter from the marketplace. This type of action will serve as a message for all peanut processors in The Philippines. The significance of increased public awareness of health hazards associated with aflatoxins in peanuts, as well as in corn and other commodities, is tremendous and long-lasting.

2. Research Capacity

Significant progress has been made in training research and support personnel in Thailand and The Philippines as well as in the U.S. since 1990. An estimated 34 undergraduate students at Kasetsart University and The University of the Philippines at Los Baños have developed thesis projects on various aspects of peanut utilization. Master of Science degrees have been awarded to 15 Thais and Filipinos at host country institutions. An additional nine Thais and Filipinos have earned or are in the process of completing M.S. and Ph.D. degrees in Food Science at the University of Georgia. Two project members from Thailand and one from The Philippines spent up to 3 months at the University of Georgia for the purpose of learning new research techniques. These instruction and training activities have resulted in a tremendous increase in personnel with a high level of knowledge and capability in peanut utilization science. The impact of these individuals on a regional level will continue long after the project ends.

GA/PH/C

Postharvest Handling Systems for the Small Peanut Producer

University of Georgia
Manjeet S. Chinnan, Ph. D.

Caribbean Agricultural Research and Development Institute
Dr. Joe Lindsay (PI-Jamaica)
Mr. A. K. Sinha (PI-Belize)

INTRODUCTION

It is a well documented fact that the quality control of farm commodities begins on the farm and continues through postharvest handling, storage and processing steps until the point of consumption. Improper handling of peanuts after they are harvested can be a primary factor associated with the reduction of market value and decrease in probability of consumption. Another factor affecting the probability of consumption of indigenously grown peanuts is their cost, which in turn is influenced by the cost associated with production, harvesting and postharvest operations.

Need for improved postharvest operations exist not only in the Caribbean region but also in the Southeast Asian countries, such as Thailand and the Philippines, and other peanut growing regions of the developing nation. Emphasis continues in the Caribbean region, whereas collaboration in Thailand was initiated in 1993-1994 and continued through June 1995. The collaboration undertaken at Khon Kaen University in conjunction with the peanut utilization project (GA/FT/TP) at Kasetsart University, Bangkok, Thailand, for an improved system of postharvest operations and providing linkage with the production and utilization aspects of peanuts. Details of the Thai collaboration component is described in the GA/FT/TP report for this year.

Overall due to very reduced level of funding activities were drastically cut. Funding for the project during this one year period was only $14,400, compared to $100,000 two years ago and $75,000 a year ago.

ACCOMPLISHMENTS

Belize

Main emphasis was on conducting training sessions for consumer taste panels and aflatoxin monitoring in peanut and peanut products. Dr. Margaret Hinds traveled to Belize for one week to conduct the training sessions. Questionnaire for conducting socio-economic studies was finalized based on that developed for the Jamaica study. Efforts continued for activities in the area of maintaining post harvest quality and reducing post production cost through improved post harvest handling operations including harvesting, threshing/stripping shelling and storage.

Jamaica

CARDI has been working very closely with UFMC (United Farmers Multi-purpose Cooperative), and has made significant progress in training of personnel and utilization of postharvest handling equipment developed or obtained through the project.

The data obtained from the socioeconomic study conducted during the last year has been fully analyzed and the report has been prepared. The main objectives of the study were: (i) Assessment of resource (land labor, off-farm income, capital) availability and utilization. (ii) Social and economic constraints at stage of production and post harvest handling operations. (iii) Cost of production, storage and disposal. (iv) Levels of technical competence, actual use of different techniques.

During the year under review several field days were held. Proportion of male and female participants was almost equal.

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US Institution - University of Georgia

The main emphasis was coordination of activities in Jamaica as well as in Belize. These activities are described under respective host country accomplishments. The work done by Dr. Hinds (Post-doc) and graduate students (Ramos and Jangchud) applicable to the U. S. food industry has been described in detail in Dr. Larry R. Beuchat's project (GA/FT/TP).

IMPACTS

Identification, modification and/or adaptation of postharvest equipment and operations to improve quality of peanuts. Medium and small scale systems such as mechanical threshers, shellers motor driven and pedal powered), mechanical dryers (stationary and portable) are available to the farmers associated with cooperatives (typically in Belize) or individual farmers (typically in Jamaica) and also to the members of UFM cooperative in Jamaica. The design of these equipment is also available so that these can be fabricated locally.

Improving quality of peanut products in host countries by working with local processors. Efforts have been made to work closely with the processors to assist in improving their peanut products. For example, in Belize these are Piache, Triple-F, and Burns; in Jamaica these are Pioneer Chocolate, and Blakey's.

Training and professional development. This is an integral part of the project both in Belize and Jamaica by working closely with BFAC and Ministry of Agricultural/Rural Agricultural Development Association, respectively.

Developing products and processes to benefit the US consumer and US food industry. Development of a prototype peanut based beverage, and edible films based on peanut proteins.. Additional funding and close cooperation with a US company should allow us to bring this project closer to commercialization.

GOAL

To develop a global model for evaluating the acceptability and profitability of peanut postharvest system(s) for Caribbean countries, in particular, and developing countries, in general, by developing and/or adapting technologies for the available economic and natural resources and by incorporating climate, socioeconomic and existing infrastructure for marketing and consumer acceptance factors.

OBJECTIVES

The proposed research is focused on attainable goals during the five year period beginning July 1, 1990. Specific objectives are:

1. Identify the segments of the postharvest handling system, and various constraints, and develop a qualitative relationship(s) among the segments.
2. Evaluate the economic value of each postharvest handling operation or segment identified.
3. Identify existing technologies for adaptation or modification.
4. Evaluate the technologies identified for functionality and economic value.
5. Evaluate the anticipated change in quality loss.
6. Determine the physical properties of peanuts and associated materials.
7. Identify the input and output parameters of all the components of the global model, and build the model.
8. Develop alternative scenarios of integrated systems.
9. Test the global model in field for functionality and acceptability.

ORGANIZATION

U. S. Lead Institution: University of Georgia

Department of Food Science and Technology, Griffin
Dr. Manjeet S. Chinnan, Principal Investigator
Mr. Glen D. Farrell, Research Coordinator I
Dr. V. M. Balasubramaniam, Postdoctoral Research Engineer

Dr. Margaret J. Hinds, Postdoctoral Research Associate

Department of Agricultural Economics, Griffin
Dr. Stanley Fletcher

Host Country Institutions:

CARDI (Caribbean Agricultural Research and Development Institute)

Belize
Mr. A. K. Sinha, Principal Investigator
Mr. Neville C. McAndrew, Agronomist
Mr. Martin Lindo, Technical Assistant
Mr. B.O. Enriquez, Manager, BFAC (Belize Federation of Agricultural Cooperative).
Mr. J. Garcia, Agronomist, BFAC.

Jamaica
Dr. Joe Lindsay, Principal Investigator
Dr. Janice Reid, Country Representative
Mr. Morris Taylor, Field Assistant
Mr. Neville Wright, Field Assistant
Mrs. C. Wilson, Extension Officer, RADA/MOA (Rural Agricultural Development Authority/Ministry Ministry of Agriculture)

ACCOMPLISHMENTS IN DETAIL

RESEARCH

A. Belize

1. SUMMARY OF ACTIVITIES DURING DR. HINDS VISIT. Objectives of the visit were:

1. Visit three local peanut processing plants to advise them on improving the quality of their products, particularly peanut butter.
2. Conduct consumer taste panels on peanut butter made in Belize and at CFSQE, the University of Georgia, from peanuts grown in Belize.
3. Hold a one-day training workshop on preparation and testing of peanuts for the presence of aflatoxin.

Objective 1. To visit with and provide technical information to local peanut processors to assist them in improving the quality of locally - manufactured peanut butter.

Presently the peanut butter produced locally is of very poor quality (viz. oil separation, lumpy texture, little or no roasted peanut flavor, noticeable off flavors e.g. caramel, oily and bitter). Purchasing and consumption of peanut butter has declined considerably because locally-manufactured brands only are available on the market. Three peanut butter manufacturers, (Piache, Triple F and Burns) were visited. Each was provided with a handout containing typical peanut butter formulations and techniques and standards for peanut butter manufacture from Woodroof's book on Peanut Processing (1983). Current operations used were discussed and specific recommendations to improve overall quality of peanut butter were made. Example of evaluation and recommendations for peanut butter manufacturing at one location (Triple F Factory, Spanish Lookout, Cayo District) is given below

a. Current process for peanut butter manufacture

Roast peanuts In shell in rotary oven (Tennessee Red and ICGV cultivars used)

|

Cool in perforated bed with fan

|

Shell and blanch in shredding machine (shells and testas are removed but seeds are not split)

|

Sort (Sorters do not remove hearts from seeds)

|

Grind peanuts in screw-propeller mill
(No heat applied; no heat control or monitoring done - actual temperature of paste is not known)

|

Add partially hydrogenated vegetable oil
Floro-Vo Brand (50 lb: 150 lb peanut paste),
stabilizer Flakex (~4%) and salt to ground peanut paste

|

Add syrup (made from white cane sugar) and mix manually

|

Fill bottles manually

|

Cool bottles at Room Temperature (80-90°F)

b. Recommendations

(i) Alterations to Process Operations

(1) Shell and grade peanuts before roasting.
(2) Remove hearts from seeds of roasted peanuts.
(3) Grind peanuts at 160-165°F in a colloid mill. Monitor temperature during grinding.
(4) Either reduce quantity of partially hydrogenated vegetable oil to ~3 % (i.e. 5 lb: 150 lb peanut paste) or use ~2% vegetable oil (e.g. corn oil or peanut oil) instead.
(5) Use granulated white cane sugar. Do not make a syrup from sugar.
(6) Do not add Flakex stabilizer at the same time with the vegetable oil and other ingredients. Stabilizer is to be added last i.e. when all other ingredients have been completely blended in the mixture. Temperature of mixture should be ~160°F when stabilizer is added.
(7) Fill bottles with mixture at ~140°F.
(8) Cool bottles in tap water for 1/2 hr. then transfer them to refrigerated storage (~50°F) for 2 days to **continue** cooling. Do **not** disturb (shake) bottled during cooling period.

(ii) To set up or be purchased:

(1) Industrial thermometers to monitor temperature of ground mixture.
(2) Colloid mill to ground peanuts.
(3) Ring burner to keep ground mixture (in mixing pail) at appropriate temperature.
(4) If shredding machine is unable to split peanut seeds or remove hearts, then a small blancher to split seeds and remove hearts **should** be purchased.

Objective 2. To conduct consumer taste panels on peanut butter samples made from Belizean-grown peanuts. Results will assist Peanut butter manufacturers in altering their formulations to make locally produced peanut butter more acceptable to Belizean consumers.

a. Peanut Butter Samples

A total of 8 sets of peanut butter samples were used. One set was a brand (Matouk's) previous imported from Trinidad to Belize and which was generally considered to be acceptable by Belizean consumers. Three sets of samples were manufactured in Belize - one from each of the peanut butter processors: Piache, Triple F and Burns. The other four sets of samples were made at CFSQE, UGA from peanuts that were obtained from CARDI, Belize. Three-digit random codes were assigned to the peanut butter samples as follows (presently, Tennessee Red is the main variety used to make peanut butter in Belize).

Code	Peanut Butter Samples
623	Matouk's (imported), variety unknown
572	Gabela (from Piache), Tennessee Red Variety
174	Triple F, Tennessee Red Variety
451	Timmys (from Burns), variety unknown
280	Made at UGA, Tennessee Red
189	Made at UGA, Tennessee Red with 2% corn oil
034	Made at UGA, ICGV88386 variety - at least 90 of these nuts were immature
322	Made at UGA ICGV88273A variety

Mr. Sinha, CARDI, was responsible for recruiting a total of 60-70 consumers from the eastern, central and western regions of Belize. Guidelines provided to Mr. Sinha included - persons invited to participate must not be allergic to peanuts; should be accustomed to eating peanut butter; should be able to read, write and fill out a simple questionnaire; should be non-smokers and not presently suffering from the 'flu' or common cold. Participants were also advised to refrain from eating spicy foods, candy or sweet beverages for one hour before coming to the test site.

b. Conducting of Taste Panels

Each panelist was provided with the 8 peanut butter samples (coded as previously explained), 8 plastic spoons, crackers and tap water to refreshen their taste buds, 1 expectorant cup, napkins, 1 pencil, questionnaire and consent forms. Consent forms were color-coded: consumers kept blue copies, CARDI kept yellow copies, the investigator (M. Hinds) kept the white copies which were attached to the questionnaires.

c. Panelists

A total of 63 persons participated: 34 of whom were 16-19 years old and the others were 20-50 years old.

d. Preliminary Findings

Overall acceptability scores for the 8 samples and comments were summarized and provided to the manufacturers. This information will assist peanut butter manufacturers in altering peanut butter formulations to make them more acceptable to Belizean consumers. Findings on four samples are tabulated below:

ATTRIBUTES	SAMPLE CODES			
	572	451	174	189
Mean scores for overall acceptability*	4.7	3.8	3.1	6.8
Liked sample(%)**	38.1	25.4	15.9	77.8
Disliked sample (%)	47.6	66.7	76.2	11.1
Would buy (%)	23.8	11.1	9.5	71.4
Would not buy (%)	47.6	63.5	77.8	4.8
Sample too sweet (%)		74.6		
Not sweet enough (%)	71.4	61.9		
Not salty enough	69.8	69.8	73.0	
Too oily (%)	65.1	47.6	68.2	
Oil just right (%)		25.4		

**Meaning of scores:
9= like extremely, 8= like very much, 7=like moderately, 6= like slightly, 5=neither /nor,
1= dislike extremely, 2= dislike very much, 3=dislike moderately, 4= dislike slightly.
** percent responded

Objective 3. To sensitize and train persons in various sectors of the Belizean peanut industry in appropriate methods for testing peanuts for aflatoxin.

Presently, only peanuts to be exported are tested for aflatoxin. Participants agreed that all peanuts should be tested. Two farmers belonging to co-operatives were concerned about the protocol to be implemented for screening of farmers' peanuts for aflatoxin. They disclosed that it is very difficult to impress upon farmers who do not belong to co-operatives about the importance of drying peanuts to appropriate moisture levels. Improperly dried peanuts are usually sold to local processors at a cheaper price than the price requested by co-operatives for properly dried peanuts. Farmers attending the workshop were concerned that a similar situation may arise if general screening of peanuts for aflatoxin were to be implemented.

A wide cross section of persons representing the local peanut industry attended the training session. Although 7 trainees had no previous laboratory experience, each participant had the opportunity to prepare samples and testing solutions and to conduct a complete Target Test on his/her own.

B. Jamaica

1. UNITED FARMERS MULTI-PURPOSE COOPERATIVE (UFMC). CARDI has been working closely with the UFMC. Although UFMC is still in its infancy, it has made steady progress since it was initiated about three years ago. CARDI has been assisting the cooperative in terms of inputs on the production of peanuts as well as postharvest handling, storage and marketing of peanuts.

Two peanut threshers operated by CARDI have been used by the cooperative members. The use of the threshers was demonstrated to three groups of farmers (about 50 persons) in several locations including Grosmond and New Hampton areas of St. Elizabeth. Subsequent to the demonstrations the threshers have been used on five

during the Spring 1995 crop. The motorized and hand operated threshers are now managed by the cooperative. A farmer has been identified to service and maintain the threshers. The UFMC arranges transport to the various farms and a small fee is charged for fuel and maintenance as well as operator's time.

In the area of marketing, the cooperative members produce the peanuts and the Co-op collects and markets in bulk. The primary outlets are Pioneer Chocolate Company, Jamaica Cereal Foods and other small processors. The cooperative is also receiving assistance in the training of members in peanut processing. Current activities include solar roasting of peanuts and making peanut candies. These products are sold through the schools in the area. CARDI has recently loaned a used commercial sealer to facilitate proper sealing and storage of processed products. In addition the UFMC is marketing its own brand of weaning food called 'Manna' which is produced by Jamaica Cereal Foods but made from peanuts and corn grown by the Co-op. A range of peanut flavored milk drinks are also being prepared by the Co-op, which is being done in a conjunction with a commercial operator of marketing milk solids.

The Co-op is in the process of obtaining the lease for a portion of land and an abandoned building for its head office as well as a factory for processing various peanut products. Substantial work has been done on the building which will be a major boost to the peanut producers in the area.

2. SOCIO-ECONOMIC STUDY. The survey was conducted in the Parish of St. Elizabeth where the bulk of peanuts are produced. Interviews were carried out in 60 peanut growing districts. The main objectives were:

1. Assessment of resource (land labor, off-farm income, capital) availability and utilization.
2. Social and economic constraints at stage of production and post harvest handling operations.
3. Cost of production, storage and disposal.
4. Levels of technical competence, actual use of different techniques.

Profile of St. Elizabeth farmer. Seventy five percent of the farmers are male. Some 70% of them live within 1.5 km of their farm where they travel mainly by walking. About 40% are over 50 years old and 33% are below 40 years of age. Some 56% completed primary school education, 27% did not complete primary school, and 10% attended secondary or higher education school.

Farm labor. The peanut farmer is generally a full time farm worker whereas 10-20% of his relatives spend 1-3 days on the time during critical times such as planting, weeding, harvesting and threshing.

Land Cultivated. The amount of land cultivated is summarized below:

Area (ha)	Percent
<0.4	10.0
0.4 to 0.8	46.5
>0.8 to 2	30.4
>2 to 4	9.0
>4 to 8	1.0

The data showed that >80 % of the farmers farmed less than 2 ha of land.

In terms of land tenure, 11.2% owned their land whereas 24.7% occupied family land. Land leased from government and other sources accounted for 34% and 16%, respectively.

Land under peanut cultivation. The data shown below indicates that more than 70% of the peanut farmers has less than one hectare of land under peanut production in any season. It is hence a typical subsistence crop.

Area (ha)	Percent
<0.4	24.0
0.4 to 0.8	49.7
>0.8 to 2	15.4
>2 to 4	1.0

Soil Tillage. Land preparation is done predominantly (75%) by tractor. However hand tools are used for tillage on 17% of the farms. Animal drawn equipment was used by 12.2% of the farmers.

Planting. It is invariably done by hand. Row planting is done on 66% of the farms whereas broadcasting is done on 24%. Broadcasting is a faster operation, the seeds are covered by a harrowing operation; however, broadcasting does not allow for molding, inter-row cultivation, and harvesting with a peanut lifter. Farmers owned 60% of the tools used out of which only 10% were power tools.

Seeds. About 80% of the farmers save their own seeds for planting. Of the remainder 10% obtained seeds from fellow farmers. Valencia was the cultivar planted on 80% of the farms. Some planted both the Valencia and CARDI-Payne, however, only 1% planted exclusively CARDI-Payne. Farmers planting CARDI-Payne grew the crop because of its marketability to the processors and taste. The primary reason for planting peanuts was for the source of income it provided.

Fertilizer use. A majority of farmers (74%) applied different grades of fertilizer to at least 0.2 ha of land. Organic matter was used by a few farmers.

Postharvest Activity. Ninety four percent of farmers harvested peanuts by hand pulling. Threshing was done by hand by the majority of the farmers also (88%). Except for the two threshers operated by Peanut-CRSP project there is only one additional thresher in the island. Threshing was considered to be one of the most arduous task in the production of peanuts. Whereas the threshers when available can handle only peanuts that have been partially dried. Due to problems with the security it is not a very attractive proposition for the average farmer.

Drying was done in the field by 52% of the farmers, sheeting on the ground was used by 23% whereas bar-be-que accounted for 3%. To reduce larceny and other losses peanuts are generally reaped, threshed and taken home for drying where there is proper supervision.

Shelling for planting is done by hand by many small farmers. From the survey 51% of the farmers do some or all shelling by hand. However, 28% used motorized sheller and 2% used a hand sheller. Some 14% used a combination of hand and motorized sheller, the shellers are private or owned or shared by a group. Some farmers felt shellers were too far from their home or farm. In fact about 50% complained of the unavailability of the sheller as a problem and 13% considered unavailability on the farm as a problem. Four percent considered the hand sheller to be difficult to operate. Some farmers refused to use the motorized sheller due to cracking of larger nuts including CARDI-Payne. The lack of electricity in some locations and the inadequate transportation system were also reported as is limitations.

Farmers were asked specific questions about the Peanut-CRSP sheller and threshers. Motorized threshers were known by 24% and pedal operated thresher by 9% of them. The hand sheller was familiar to 7% of the farmers interviewed. However, 24% of the farmers did not know about CARDI and 23% did not know about the peanut equipment evaluated by CARDI. Farmers were willing to pay to have their peanuts threshed, 38% would use equipment if they are placed at a central location.

Bags were used for storage receptacles for peanuts on 80% of farms. Various places were used for storing peanuts, such as special storage room (59%), kitchen (29%), cellar (2%), community storage (1%), other (9%).

The farmers interviewed reported a number of postharvest problems. These are - marketing(33%), removal from field (17%), thresh and pick (12%), labor availability (9%), theft/larceny (4%), drying (4%) and pests (4%).

In case of crop protection 5% of farmers took precautions against rats. Such precautions include the use of rat baits (64%) and keeping a cat (7%). Precautions against include proper drying (43%), removal damaged and infested material (28%) and airing at periodic intervals. Persons taking no special precautions made up of 27%. Removal of damaged pods was used to control molds.

Cooperatives. The farmers in general were not too eager to form co-ops. Although 25% considered co-ops to be useful, 5% believed that they will never work, 7% considered only some benefit. Marketing cooperatives were considered to be worthwhile by less than 2%.

C. U. S. Institution - University of Georgia

The main emphasis was coordination of activities in Jamaica as well as in Belize. These activities are described under respective host country accomplishments. The work done by Dr. Hinds (Post-doc) and graduate students (Ramos and Jangchud) applicable to the U. S. food industry has been described in detail in Dr. Larry R. Beuchat's project (GA/FT/TP).

TRAINING

CARDI-Jamaica Personnel: Morris Taylor, Dr. Joseph Lindsay and Carol Wilson offered technical assistance and training as needed to peanut growers in Jamaica.

CARDI-Belize Personnel: Dr. Margaret Hinds traveled to Belize to provide training on various aspects of postharvest handling, product development and quality evaluation. Members of several cooperatives associated with BFAC received lectures on production and postharvest handling of peanuts. A number of field days were organized jointly with BFAC.

INTERNATIONAL TRAVEL

Dr. Margaret J. Hinds traveled to Belize from September 5 to 10, 1994 to hold a training session for conducting consumer tests, aflatoxin monitoring, and advising local processors to improve quality of their peanut products.

Dr. Joe Lindsay from CARDI-Jamaica traveled to Washington D.C. to participate in the International Research Symposium and Workshop organized by Peanut CRSP, March 29-31, 1995.

Mr. Anil Sinha from CARDI-Belize traveled to Washington D.C. to participate in the International Research Symposium and Workshop organized by Peanut CRSP, March 29-31, 1995.

PUBLICATIONS AND PRESENTATIONS

Chinnan, M. S. 1995. Improving peanut postharvest systems. Paper presented at the Peanut-CRSP Workshop and Symposium on Impacts and Scientific Advances through Collaborative Research on Peanuts. March 29-31. Washington, D. C.

Hinds, M. J. 1995. Quality enhancement and technology transfer of peanut butter. Paper presented at the Peanut-CRSP Workshop and Symposium on Impacts and Scientific Advances through Collaborative Research on Peanuts. March 29-31. Washington, D. C.

Lindsay, J. I. 1995. CARDI-Payne extension and impacts in Jamaica. Paper presented at the Peanut-CRSP Workshop and Symposium on Impacts and Scientific Advances through Collaborative Research on Peanuts. March 29-31. Washington, D. C.

Sinha, A. 1995. Peanut farmer cooperatives in Belize. Paper presented at the Peanut-CRSP Workshop and Symposium on Impacts and Scientific Advances through Collaborative Research on Peanuts. March 29-31. Washington, D. C.

Hinds, M. J., Beuchat, L. R. and Chinnan, M. S. 1995. Particle size distribution in a peanut based beverage. (abstract) American Peanut Research and Education Society Meeting, Tulsa, OK, July 12-14, p 77.

Hinds, M. J., Chinnan, M. S. and Beuchat, L. R. 1995. Acceptance and purchase intent of peanut butter by Belizean consumers (Abstract) Annual Meeting of the Institute of Food Technologists, Anaheim, CA, June 3-7, Book of Abstracts, p 230.

PLANS FOR 1995-1996

1. Research thrusts/endeavors planned:

University of Georgia:

A. Postharvest handling system. Coordination of activities in Jamaica and Belize will continue to obtain additional data on postharvest handling systems. Will also serve as linkage to other countries in the region as needed.

B. Aflatoxin testing. Aflatoxin levels on selected samples of peanuts in grown in Belize and Jamaica will be tested at CFSQE.

C. Drying system. Develop a computer model for designing a drying system for different energy sources.

D. Utilization of Peanuts. Alternative processing approaches to improve the quality of food products made from Caribbean grown peanuts will be studied. Emphasis will be given on improving the stability and quality of indigenously manufactured peanut products.

Host Country

Jamaica

A. Evaluation of peanut threshers The Thai thresher will be further tested and demonstrated to the local farmers.

B. Cooperation with a local college Cooperation with the College of Arts Science and Technology (CAST) on modification/construction of mechanical aids will continue.

C. Socioeconomic survey Data analysis and the final report on the socioeconomic-economic study will be completed.

Belize

A. Socioeconomic survey. Socioeconomic survey instrument has been modified to be used for conducting a study. The data will be collected and analyzed.

B. Peanut Thresher. A small peanut thresher imported from Khon Kaen (Thailand) will be further tested for the conditions in Belize.

C. Peanut Dryer. Work on testing of the peanut dryers will continue.

2. Training Planned

Training of one CARDI and one BFAC person to obtain socioeconomic data will be done.

Dr. Margaret Hinds, will visit Jamaica to train CARDI personnel on the use of VICAM system for conducting aflatoxin analysis and assisting local processors in improving quality of value-added peanut products.

3. Manuscripts etc.

Manuscripts (1-2) will be written and submitted for publication in refereed scientific journals.
Presentations (2-3) will be given at professional scientific meetings.

4. International Travel

One trip for U.S. PI to Host Country for project review, and one trip for Dr. Margaret Hinds to travel to host country (Jamaica) to conduct training sessions.

5. Timetable.

All activities will be in progress throughout the project period.