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National Program 301: Plant, Microbial and Insect Genetic Resources, Genomics & Genetic Improvement

Accomplishment Report 2000-2005

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BACKGROUND AND GENERAL INFORMATION

U.S. systems of renewable resource production and land stewardship face formidable challenges, among the most exacting of which is successfully adapting to the accelerating rates of change in factors affecting agricultural productivity. The demands placed on the national system of renewable resource production by a rapidly changing world can only be met by technologies that optimally harness the inherent genetic potential of plant, microbial, and insect germplasm. Production systems that optimally preserve and harness that genetic potential will maximize profits, security of supply, price stability, market competitiveness, and avoid crop losses from genetic vulnerability.

The vision for the USDA/ARS National Program 301 Plant, Microbial, and Insect Genetic Resources, Genomics, and Genetic Improvement is to furnish genetic and bioinformatic tools, genomic information, and genetic raw materials to enhance U.S. agricultural productivity to ensure a high quality, safe supply of food, fiber, feed, ornamentals, and industrial products. Consequently, its mission is to safeguard and utilize plant, microbial, and insect genetic resources, associated genetic and genomic databases, and bioinformatic tools to ensure an abundant, safe, and inexpensive supply of such products for the United States and other nations. This National Program's mission follows from the USDA/Agricultural Research Service Strategic Plan (see http://www.ars.usda.gov/aboutus/docs.htm?docid=1766) which, in turn, is directed towards achieving goals mandated by the USDA Research, Education, and Extension Mission Area Strategic Plan and the USDA Strategic Plan for 2002-2007 (see http://www.usda.gov/ocfo/usdasp/usdasp.htm).

The products of NP 301's research contribute toward broader goals (termed "Actionable Strategies") associated with two specific Performance Measures from the ARS Strategic Plan for 2003-2007 Goal 1: Enhance Economic Opportunities for Agricultural Producers.

Performance Measure 1.2.7**:** Identify genes responsible for plant product quality and resistance to disease, pests, and weather losses. Target: Have a more complete understanding of the genes responsible for quality, growth, and health of agronomic crop species.

Performance Measure 1.2.8: Maintain, characterize, and use genetic resources to optimize, safeguard, and enhance genetic diversity and promote viable and vigorous plant production systems. Target: The diversity of the germplasm collections will be expanded by acquisition of new accessions; and genetic resources from these collections will be used to produce new and improved food, agricultural, and industrial applications for agricultural products.

PLANNING AND COORDINATION FOR NP 301

USDA/ARS National Programs follow a five-year program cycle, initiated by a Customer/Stakeholder Workshop. The first NP 301 National Program Cycle began with a workshop in April, 2000, in Atlanta, Georgia. ARS scientists and administrators met with customers, stakeholders, and partners and discussed major crop agricultural issues and research priorities. Based on these in-depth discussions, major Research Components for this National

Program (NP) were identified, prior to developing the NP 301 Action Plan (see http://www.ars.usda.gov/research/programs/programs.htm?np_code=301&docid=1013).

The NP 301 Action Plan was drafted by writing teams composed of ARS scientists and members of the USDA/ARS National Program Staff (NPS). The writing teams combined input from the workshop, their own knowledge of the subject matter area, and input from other ARS scientists and their cooperators to identify the key, priority needs that could be addressed by ARS research. These needs were aggregated into Problem Areas for each NP Research Component. After a public comment period, the draft Action Plan was revised and completed in 2001.

Once the Action Plan was completed, specific research Project Plans were written by individual scientists or teams of scientists. Project Plans included statements of the anticipated products or information to be generated by the Project, how they contributed to solving the larger National Program Problem Areas, and time lines and milestones for measuring progress toward achieving the Project goals. All Projects Plans associated with NP 301 were then evaluated for scientific quality by external peer panels. The project peer reviews were handled by the ARS Office of Scientific Quality Review. Project Plans were revised in response to review panel recommendations, and then implemented. Five years since the first NP 301 Customer-Stakeholder Workshop in 2000, the progress achieved in attaining the Action Plan goals is now being assessed by an external assessment panel. This assessment is in preparation for the beginning of the next five-year National Program Cycle.

NP 301 is ARS's largest National Program and, as such, requires ongoing coordination at the national level. Such day-to-day coordination is the task of National Program Leaders who comprise the NP 301 Leadership Team. USDA/ARS National Programs are also coordinated with other ARS National Programs and with activities of other agencies. For example, discussions and analyses of the Interagency Working Groups (IWGs) on Plant Genomes and Microbial Genomes coordinate and align NP 301 plant and microbial genome research with efforts in other agencies. The IWGs include representatives from the USDA (ARS and CSREES, Cooperative State Research Education and Extension Service), National Science Foundation (NSF), Department of Energy (DOE), Office of Science and Technology Policy (OSTP), and the Office of Management and Budget (OMB).

In addition to ongoing planning and coordination during the first NP 301 Program Cycle 2000-2005, USDA/ARS National Program Staff and Area Offices organized and conducted many workshops focused on specific research issues relevant to the needs of U.S. agriculture. Most of these workshops also involved coordinating and integrating ARS NP 301 efforts with those of cooperating agencies, and with university and industry partners. A partial list of workshops related to NP 301 appears in **Appendix 1 – Selected Supporting Information and Documentation for Accomplishments and Impact of NP301 Research** (1).

HOW THIS REPORT WAS CONSTRUCTED AND WHAT IT REFLECTS

In this Report, information about National Program 301 achievements and their impact is organized according to three National Program Research Components and their constituent Problem Areas, described in the National Program Action Plan. The report first outlines the

three NP 301 Research Components and the **outcomes, goals and commitments** for Problem Areas within each Component. These are followed by selected accomplishments achieved during the last five years and by the **impact** of those achievements on solving the problems and meeting the high priority needs identified by customer/stakeholders in the NP301 Action Plan.

For the most part, the content of this report is derived from responses to a recent survey of the scientists assigned to NP 301, who were asked to summarize their project's major accomplishment during the last five years, its impact, and key references documenting that accomplishment. Consequently, this report does **not** include **all** accomplishments achieved by each NP 301 research project but, rather, only those **selected** by the ARS scientists polled, and the ARS scientists and National Program Leaders who authored this report. As a result, the scope of this report encompasses a subset of the total spectrum of NP 301 accomplishments, chosen to illustrate and exemplify the total progress and achievements of this large National Program.

NP 301 encompasses more than 180 research projects. The titles of the individual projects are listed in **Appendix 2 – Research Projects**, which is organized according to the relevant NP 301 Research Component and geographical location of the research unit. Note from Appendix 2 that the individual research projects have not been associated with specific Problem Areas because, in general, those projects address the challenges and problems of multiple Problem Areas. Also note that some of these projects began more recently than the initiation of the first NP 301 National Program Cycle five years ago.

NP 301 Research Components I and III address, respectively, genetic resource management and genome databases and bioinformatics. Those Research Components include fewer individual projects than does Research Component II, which focuses on plant genetics/genomics and breeding. Furthermore, applied research and service, rather than "discovery research" per se, is the primary focus for relatively many projects assigned to Research Components I and III. These differences, among Research Components I and III, relative to Component II, are evident in the project lists in Appendix 2, and should be borne in mind while reading this report. Similarly, only a few NP 301 projects address microbial or insect germplasm, genomics, or genetics, so this Report emphasizes plant-related research and service efforts.

Finally, a word about how NP 301 achievements were documented. Just as only selected accomplishments are reported, the details of those accomplishments are documented selectively so as to illustrate the overall variety of products and knowledge generated by this National Program. In the report text, selected assertions found in the narrative are cross-referenced, by numerical citation [e.g., "(1)"], to supporting information presented in **Appendix 1.** Appendix 1 is organized according to the ten NP 301 Problem Areas, and also includes a list of workshops which were conducted during 2000-2005 to address specific NP 301 programmatic issues.

RESEARCH COMPONENT I: GENETIC RESOURCE MANAGEMENT

Five years ago, this National Program Component committed to the goals that microbial and insect germplasm should be acquired and safeguarded through the expansion of genebanks or *in situ* preserves for long-term accessibility. The genetic content of acquired germplasm should be characterized to insure broad-spectrum genetic variability while minimizing genetic redundancy within genebank collections. The agricultural potential of unimproved germplasm must be evaluated comprehensively.

The following summaries, organized by Problem Areas, describe the goals that this Research Component addressed, and progress achieved in attaining those goals. In these summaries, and throughout the report, "germplasm" is used synonymously with "genetic resources." Conversely, "evaluation" and "characterization" are not considered synonyms herein. Evaluation involves assessing agronomic or horticultural merit, generally through analyzing variability in traits with complicated patterns of inheritance and significant genotype by environment interactions. Evaluations generally require testing over multiple test environments. In contrast, characterization involves assaying more simply inherited traits (e.g., molecular or color variants), often with discrete character states, and no or minimal genotype by environment interactions. Characterizations are generally conducted to assess genetic variability in genetic resources, to identify unknown germplasm samples, and to elucidate systematic and genetic interrelationships.

Problem Area Ia – Safeguarding Threatened Genetic Resources and Associated Information

Outcomes, Goals, and Commitments: Critical goals for this Problem Area are to identify and fill gaps in the genetic diversity (and in basic biological knowledge about that diversity) preserved in USDA/ARS plant, microbial, and insect germplasm collections. To fill those gaps, new germplasm samples must be acquired, either by conducting foreign or domestic explorations, or by interinstitutional exchange. International germplasm exploration or exchange often first involves establishing international agreements with other nations; in some cases, the USDA/ARS may agree to support genetic resource management in donor nations. Once new samples of genetic resources or associated information are acquired, they are incorporated into USDA/ARS collections and associated databases; some samples must first be processed through quarantine facilities. This Problem Area not only seeks to safeguard threatened genetic resources and associated information, but also to develop and implement new or improved scientific strategies, methods or technologies for accomplishing that preceding goal.

Selected Accomplishments: During the last five years, gaps or under-representation of genetic diversity in U.S. collections were identified by curators and filled through acquisition of genetic resources. Seventy-two plant explorations and exchanges to acquire landraces and wild relatives of crops were funded and coordinated in the United States and 25 foreign countries on four continents (1,2). NP 301 scientists collaborated with other U.S. and international donor agencies, international research organizations, CGIAR centers, and national research and development programs in 23 countries to support the conservation and exchange of genetic resources (3). This support included establishing innovative capacity-building partnerships to assist countries in renovating and upgrading their genebanks and training their personnel.

During this period, more than 5,000 plant germplasm accessions were acquired through plant explorations by the U.S. National Plant Germplasm System (NPGS), spanning the full spectrum of U.S. major and minor crops (4), e.g., more than 800 new accessions of fruits, nuts, and specialty crops. Those acquisitions preserved landraces and wild germplasm that are rapidly disappearing. As that germplasm disappears from the field, the accessions preserved *ex situ* may become the only surviving source of that genetic diversity. For example, cotton landraces in Mexico now often survive only as curiosities in garden plots and occasional feral plants. Several explorations to Mexico resulted in the acquisition of numerous cotton landraces and a new *Gossypium* species.

The joint ARS-APHIS plant quarantine program provided a safe mechanism for introducing germplasm into the United States. For example, the sugar cane quarantine program processed through quarantine 79 unique foreign sugar cane accessions and facilitated the interstate exchange of 99 sugarcane clones for breeders and other scientists. New pathogen tests for fruit crops were implemented to improve virus detection methods so as to satisfy foreign quarantine regulations. Across all crops, this interagency effort processed more than 12,360 germplasm samples from 46 countries to fill gaps in genetic diversity in U.S. collections.

NP 301 scientists also acquired microbial and insect germplasm critical to U.S. agriculture, and devised new technologies that facilitated that acquisition. Furthermore, studies of a large diverse collection of exotic bacterial and viral pathogens determined that a citrus pathogen, *Xylella fastidiosa*, which causes citrus variegated chlorosis disease, can be transmitted through seed (5). s a result, quarantine testing protocols for citrus seed exchange now include assays for this pathogen.

Finally, accurate and complete information about the ecogeographical origin of germplasm collections is critical for optimal use in research in breeding. During the last five years, global positioning systems (GPS) were adapted as standard equipment for all NPGS-supported plant explorations, so as to ensure that geospatial information (latitude, longitude, altitude) was recorded for newly-collected samples.

> **Impact:** Current and future improvement of U.S. crops relies on the ability to introgress new genetic variability from wild germplasm or landraces. The more than 5,000 additional plant genetic resource samples acquired through plant exploration and exchange programs in the last five years furnished U.S. scientists access to novel, genetically-diverse germplasm for genetic improvement and scientific research. Genetic resources containing novel traits from these acquisitions are now available for incorporation into elite lines. Plant genetic resources were acquired for improving most of the major crops, as well as many minor or niche crops, which will support rural development and crop diversification for small and large farming operations.
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> The germplasm quarantine program furnished a safe, secure avenue for intercepting infected material and distributing only healthy germplasm. Both known and previously unknown pathogens were identified and testing procedures have been implemented to reduce the probability that these pathogens will affect U.S. crop production.

International genetic resource conservation efforts were supported and foreign scientists were granted unrestricted access to the comprehensive U. S. germplasm collections. This has helped maintain some reciprocal access to genetic resources in other nations or in international institutions in a time when there is considerable political pressure to restrict such access.

Problem Area 1b – Conserving Genetic Resources and Associated Information Efficiently and Effectively

Outcomes, Goals, and Commitments: USDA/ARS has made a long-term commitment to maintain genetic resources effectively in genebanks and, in cooperation with other agencies or organizations, to encourage their long-term protection *in situ* in reserves. To do so also requires a long-term commitment to maintain and develop specialized facilities, infrastructure, and trained staff needed for this purpose. As new demands and conditions arise, new genebanks may be required. Duplicate, back-up samples of the collections should be maintained in more than one location to prevent catastrophic loss of valuable genetic resources.

The USDA/ARS genebanks not only strive to conserve genetic resources and associated information, but also to develop and implement new or improved curatorial strategies, methods or technologies. Specifically, superior methods are developed for estimating optimal population sample sizes for genebank collections, and optimal methods and conditions are devised for preserving genetic resources over the long-term. New and more effective methods are developed and implemented for regenerating genetic resources, and for monitoring and testing them for vigor, health, and trueness-to-type. From an operational standpoint, genetic resource accessions are effectively regenerated by genebanks from material in storage or in the field, thereby affording material for distribution to researchers. When major problems in conserving and maintaining genetic resources are solved and the solutions adopted by other genebanks and research programs, scientific or economic advantages may result.

Selected Accomplishments: During the last five years, a broad range of ARS plant, microbial and insect genetic resources were preserved in USDA/ARS genebanks under optimal maintenance conditions. The size of the NPGS grew from ca. 440,000 accessions to more than 464,000 samples at present (1). New demands, priorities, and needs for genetic resource conservation led to the establishment of several new genebank sites. Within the last five years, the Ornamental Plant Germplasm Center at Ohio State University was established, with ARS funding and direction, to maintain diverse herbaceous ornamental species that formerly had not been actively managed. Similarly, the USDA/ARS National Arctic Plant Genetic Resources Unit was established at Palmer, Alaska, to preserve arctic, sub-arctic, and alpine crop species. The recently-established National Arid Land Plant Genetic Resource Unit at Parlier, California, evolved into the priority regeneration site for plant genetic resources adapted to warm long-season and arid conditions. Other NPGS genebanks now regularly transmit relevant materials to Parlier and Palmer to be regenerated under their special field conditions.

Storage under reduced temperature conditions is vital for long-term germplasm maintenance. The USDA/ARS National Center for Genetic Resources Preservation (NCGRP) at Ft. Collins,

Colorado, added more than 70,000 duplicate plant samples in the last five years, serving effectively as a long-term, high security, safety backup for more than 80% (as compared to ca. 65% five years ago) of the entire NPGS collection, and also safely housing backup collections for major international genebanks of maize, wheat, rice and their wild relatives. Safety backup collections were also effectively maintained at other NPGS sites. For example, the genebanks in Hawaii and Puerto Rico maintained duplicate orchard plantings of the cacao collection from Miami, as part of a more general conservation effort for almost a thousand samples of diverse tropical crops grown not only in orchards, but also backed-up in tissue culture.

For agriculturally-important microbes, ARS curated the National Rhizobium Germplasm Resource collection (2) thereby maintaining symbiotic nitrogen-fixing bacteria that are major contributors to soybean productivity. Also, important cultures of insect-associated fungi from an endangered Australian culture collection were duplicated for safety in the U.S. collection for these fungi. Transgenic, mutant, and factory colonies of insect embryos from the USDA/ARS screwworm eradication program were cryopreserved for use by future generations of researchers.

In addition to expanding ARS genetic resource management capacity, NP 301 developed and implemented new or improved curatorial strategies, techniques or methods. For example, ARS scientists developed more efficient ways to preserve wild relatives of crops through novel storage regimens that incorporated new molecular and biophysical approaches. Surveys of survival following different preservation techniques led to the development and implementation of more effective storage protocols for crops and their wild relatives (3).

Superior *in vitro* culture and cryopreservation technologies were developed and applied to a variety of plant genera, enabling effective storage of new important clonal and seed genetic resources (4). Tools to quantify and apportion genetic diversity were applied to apple, garlic, hops, beet, rye, and wild rice, with the result that curators can better compare the costs of maintaining individual accessions with their respective contribution to the total genetic diversity included in collections.

For ARS insect genetic resource collections, development of better collection and storage methods for honey bee semen not only facilitated curation, but also enabled bee breeders and researchers to schedule bee breeding activities more flexibly. In addition, cryopreservation protocols were developed and successfully applied to long-term storage of insect strains for research and control programs (5). Similarly, key new discoveries about the biology, identification, and genomics of insect-associated fungi resulted in improved techniques for collecting, identifying, isolating, and preserving those fungi.

New and more effective methods for monitoring and testing genetic resources for vigor, health, and genetic identity were developed and implemented. These advances not only benefited germplasm curation, but also assisted research and breeding programs. For example, breeding and propagation of a woody ornamental species of *Halesia* were accelerated by an improved seed germination method that reduced germination time from eight months to four months.

Germination, disease, and quality testing were applied to assess the relative vigor and health of seed samples, and the need for prompt regeneration. Quality assurance capacity was expanded

significantly, with new germination testing programs established at two NPGS genebanks, and new molecular diagnostic capabilities developed at four other sites. Continual monitoring of plant germplasm health generated new reports for diseases in garlic, onion, lentil, pea, various grasses, and other crops. During the last five years, the NCGRP conducted more 80,000 germination tests on incoming back-up samples and more than 20,000 germination tests on existing accessions. Other NPGS genebanks conducted viability or germination tests on more than 50,000 samples. Similarly, with microbial germplasm, rigorous quality control testing was conducted before adding new strains to the National Rhizobium Germplasm Resource collection.

During the last five years, more than 80,000 seed-propagated accessions (about 17% of the total NPGS collection) were regenerated, an increase in "grow-out" capacity of at least 3%. Concomitantly, NPGS genebanks propagated about 20,000 clonally-reproducing accessions. Such successful, high-quality regenerations resulted in improved germplasm availability for users. For example, by regenerating accessions with low seed supply or viability, more than 2,000 vegetable accessions unavailable for distribution five years ago are now available to users.

More effective and efficient regeneration methods were developed and implemented to produce high-quality seed samples, and maintain their genetic integrity. During the last five years, controlled-pollination regeneration capabilities were established at five NPGS genebanks. For example, for forage legumes, techniques for cage-isolated bee pollination were refined; cultivation techniques were changed from direct seeding to greenhouse transplants. *In vitro* techniques to rescue vulnerable seed lots were developed, as were improved protocols for producing seeds from wild species. All of the preceding accomplishments contributed to improved quality and success of germplasm regeneration.

> **Impact:** Success with attaining this Problem Area's overall goals of conserving and making readily available superior genetic resources has spawned or catalyzed successful outcomes in essentially all NP 301 Problem Areas. Consequently, the specific impacts cited here could readily be attributed to other NP 301 Problem Areas and Research Components—an anticipated result, considering NP 301's integrated programmatic organization. To cite just a few examples, genetic resources maintained by NPGS genebanks donated biotic and abiotic stress resistance, quality, adaptation, and yield improvements to many vegetable crops. Specifically, ready access to potato landraces and wild relatives was the basis for the preceding genetic material appearing in the pedigrees of most of the four to ten new "elite" potato clones released each year. Maize genetic stocks conserved by USDA/ARS and the University of Illinois furnished genetic tools critical to the rich understanding of maize's biology and the concomitant spectacularly rapid agronomic improvements in this crop (6). Similarly, morphological descriptions of the model legume *Medicago truncatula* included in the Germplasm Resources Information Network (GRIN) benefited genomics research with this species. Finally, a strain from the National Rhizobium Germplasm Resource furnished the DNA analyzed to assemble the complete genome sequence of *Bradyrhizobium japonicum* (7).
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> During the last five years, researchers and genebank curators developed and implemented more efficient and effective genetic resource storage protocols, viability testing, regeneration procedures, core subsets, and means for estimating the incremental increase

in genetic diversity conserved by increased allocation of curatorial effort. Notably, significant improvements in long-term storage procedures and protocols for maintaining genetic resources are often incremental, and the impacts are perhaps difficult to gauge over only five years. Nevertheless, these incremental improvements are responsible for the availability of more and higher quality genetic resources. For example, identifying mislabeled and unnecessarily duplicated accessions will help users avoid conducting research on redundant or irrelevant material. Access to more detailed information about the genetic diversity residing within genebank collections may accelerate research by enabling scientists to efficiently select accessions most likely to have specific traits of interest. More accurate and detailed characterization of insect-associated fungi has answered major systematic questions about these fungi and helped researchers identify germplasm potentially valuable for insect biological control efforts. Similarly, characterizing the genetic diversity of fungal endophytes within grass accessions may contribute to grass cultivar development and research.

Problem Area Ic - Documenting and Characterizing Genetic Resources

Outcomes, Goals, and Commitments: USDA/ARS genebanks are committed to thoroughly document key genetic resource collection management and descriptive (especially passport) data in public databases, routinely updating that information to meet needs of genebank managers and scientific clientele. Database security must be continually enhanced. Coordination and linkage between accession-based genetic resource databases and genome databases--based on the characteristics and physical location of genes--are developed, maintained, and enhanced. Not only must databases and information in general be managed effectively, but new standard "user-friendly" descriptive terms and formats (descriptors) must be developed to communicate salient information in a standardized form.

Selected Accomplishments: Up-to-date documentation and timely description of crop, microbial, and insect germplasm provide curators and users with information critical for its effective conservation and expanded utilization. In the United States, the primary database for plant genetic resource management and use is GRIN, which was developed and maintained by scientists in NP 301 (1). During the first NP Cycle, GRIN was routinely updated, so that current passport and characterization/evaluation information were accessible continuously to NPGS curators, managers and the germplasm user community. New descriptors and new categories of data were regularly incorporated into the GRIN and linkages were forged to other genetic resource and genomics databases, e.g., SINGER (2). During the last five years, "updating the GRIN database" involved incorporating literally millions of new data points (3). More than 70,000 digital images are now delivered by GRIN servers.

Because of its easy access and reliability, GRIN was consulted by an extremely broad spectrum of researchers and the general public during the last five years. For example, each day during 2004 the GRIN web site was queried by an average of 200 different individual users (or their computers). Furthermore, the information needs of the NPGS genebanks and germplasm users were regularly assessed and the GRIN application software was enhanced to meet changing user needs and to ensure its compatibility with a variety of computer operating systems. To meet those needs, two major GRIN software upgrades were made. A new computer server and more

data storage and back-up hardware were procured. Effective protective measures were implemented to ensure that GRIN was shielded from malicious attacks and accidental data loss.

Nearly all NPGS genebanks are now "active GRIN nodes" that continually update GRIN information in "real time." For example, during the last five years, the Ames, Iowa, genebank added an average of ca. 32,000 observations each year to GRIN. The Griffin, Georgia, genebank routinely updated crop-specific information in GRIN with more than, more than 584,000 records modified, and more than 10,000 images added to the GRIN (4).

As needed, GRIN and other NPGS personnel trained foreign curators and data managers so as to encourage sharing of information and to optimize interoperability of genetic resource databases globally. As a result, the PC version of GRIN (pcGRIN) is now used by genetic resource managers in more than 35 nations. The 40 NPGS CGCs actively interacted with curators to improve documentation and characterization of commodity-specific NPGS collections.

Accurate identification of germplasm accessions and use of their correct scientific names are essential for effective genetic resource management and use. Online taxonomic resources key for determining the correct scientific name for specimens were provided by the USDA/ARS Germplasm Resources Information Network (GRIN), which now includes more than 28,000 verified taxonomy records which are accessible on the Internet via a completely upgraded GRIN taxonomy web interface (5). Similarly, an online database containing key fruit and seed characteristics for all legume genera was developed and now assists users worldwide in legume genera identification. Finally, NP 301 scientists collaborated in an international consortium, which included the CGIAR system, to develop online molecular and phenotypic databases for coffee and cacao.

NP 301 made significant progress with applying genetic characterization to clarifying genetic relationship among crop species and their wild relatives. For example, new integrated morphological and molecular datasets clarified the taxonomy of wild and cultivated potatoes and tomatoes. The datasets form part of an authoritative monograph that documented the history of potato germplasm collecting and taxonomic research, produced a formal synonymy of taxonomic names, and delivered botanical keys, morphological descriptions and illustrations, accounts of systematic interrelationships, detailed locality maps, geographic information systems (GIS)-based analyses of diversity, and listings of all locality records for specific regions (6).

There were similar advances for microbial genetic resource collections. For example, taxonomic keys and descriptions were constructed for the fungal associates and pathogens *Alternaria* and *Cladosporium* that clearly distinguish relevant fungal species, and now serve as important reference tools for a variety of users. Studies of microbes associated with legume genebank collections identified a pea disease with effects that mimic fungicide contamination. After reporting these findings to the USDA Federal Grain Inspection Service, pea shipments with the disease were no longer impounded because of suspected contamination.

> **Impact:** Information systems tailored to genetic resource management now ensure that passport and characterization data are readily accessible for U.S. germplasm collections. The GRIN database, developed and maintained by NP 301 scientists, comprehensively

documents the more than 460,000 NPGS accessions, enabling NPGS managers and curators to effectively manage the collections. For example, the ready access to passport information for many accessions was critical for developing optimal germplasm regeneration and long-term storage approaches. Linkages and interoperability between the GRIN and other domestic and international germplasm databases ensured that innovations in managing observation/evaluation data were widely disseminated. This information made the accessions more useful to researchers and breeders of new cultivars. Also, methods for accessing, manipulating and analyzing data about NPGS accessions were regularly improved, which enhanced the utility of the information in the GRIN database.

Safeguarding and enabling ready access to information associated with genetic resources increases their utility for research. With more than 150,000 "taxon reports" issued via the Internet each month to agricultural researchers worldwide, GRIN taxonomic data have served as the international standard source of taxonomic and nomenclatural information on the economic plants of the world.

Among other impacts, the expanded germplasm characterization and passport data have furnished users with important information about genetic and systematic relationships. For example, the greatly enhanced knowledge of the systematic relationships of potatoes and tomatoes totally revised current understanding of the number of potato and tomato species, and their interrelationships, thereby completely updating the scientific names of these important crops (7). Passport information in GRIN, such as maps and locality records, served as critical references for further germplasm collections. Improved taxonomic information, when combined with evaluation data from subsets of the entire collection, helped breeders choose the best germplasm for their breeding programs and avoid suboptimal germplasm.

Problem Area Id - Expanding Germplasm Evaluations and Characterizations

Outcomes, Goals, and Commitments: Genetic resources in USDA/ARS genebanks must be characterized for critical descriptors, and genetic variation must be estimated within and among accessions, populations, species, and/or genera. Knowledge of genetic profiles for specific genetic resources is then applied to identify gaps and duplicates in collections, and to define core subsets. Characterization and/or evaluation data must be consolidated from the literature and/or from unpublished notes, and incorporated into public databases accessible online, e.g., GenBank, GRIN, Soybase so that they are readily accessible for accelerating research and breeding. This Problem Area's goals complement those of other NP 301 Problem Areas, particularly Problem Areas IIa and IIIc.

For genetic resources to be exploited effectively by researchers and breeders, they should be evaluated across multiple years and test environments to expand knowledge of agronomic and horticultural properties. For crops, host-plant resistance to abiotic (i.e., environmental extremes) and biotic (i.e., diseases, pests) stresses are perhaps the most important attributes of genetic resources for crop improvement. But, end-use traits, such as product quality, are increasingly significant, especially for specialty (often horticultural crops), and must also be evaluated. Given

the multitude of agriculturally-important traits, important goals for this Problem Area include the development and implementation of superior methods and approaches for evaluating genetic resources more efficiently and effectively.

Selected Accomplishments: During the last five years, ARS scientists made significant progress in evaluating and characterizing germplasm for critical traits and estimating the level of trait variation in NPGS collections. More than 110,000 different accessions (about a quarter of the entire NPGS collection) were evaluated for their horticultural or agronomic merit. New crop evaluation projects were begun at five NPGS genebanks. Molecular marker capabilities were enhanced significantly at 11 NPGS sites, so that more than 75% of NPGS genebanks now have in-house molecular marker capabilities. More than 3,000 accessions were genotyped with molecular makers, and more than 10,000 were characterized morphologically. Genetic diversity in the avocado, cocoa, grape, papaya, pecan, *Prunus* and medicinal plant germplasm collections was characterized with molecular makers to enable accurate taxonomic identification and to investigate the genetic variation in the NPGS and several university collections (1-5). These data helped identify gaps and duplicates in collections, circumscribe core subsets, and identify and correct taxonomic errors.

Despite the preceding progress, with the sizable growth in the size of ARS genebank collections, the challenge of providing even the most basic descriptive data for new samples was daunting. To address that challenge, whenever genetic resources were regenerated, key traits were evaluated by curators, sometimes in concert with Crop Germplasm Committees (CGCs) and other crop specialists (6). Evaluation data were analyzed, consolidated, and deposited primarily into GRIN, but also sometimes in crop-specific, genomic/bioinformatic databases like Soybase and GenBank (see Research Component III).

Fruit tree genetic resources from the NPGS were widely exploited by public breeding programs, but their large size and perennial growth habit complicated evaluation of new accessions. Nevertheless, host-plant resistances to pear scab and pear powdery mildew were identified during extensive evaluations of the pear collection. Horticultural traits of tropical fruits were evaluated over several environments, stressing important production and consumer factors such as yield, fruit shape, and sweetness of carambola, papaya, mango, cocoa, and banana (7). Newly acquired samples of a wild progenitor species of apple were evaluated for disease and pest resistance, environmental stress tolerance, plant stature and molecular diversity in a collaborative project spearheaded by NP 301 scientists. Superior disease resistance was identified that promises to have major impact on the apple industry. This unadapted germplasm was also intercrossed with a standard well-adapted U.S. apple cultivar to accelerate incorporating this new germplasm into breeding programs (8, see also NP 301 Research Component IIa).

Similarly, landscape tree and shrub germplasm utilization was limited historically because crossability of unimproved materials with adapted commercial plants was poorly known. This crossability was characterized and horticultural traits evaluated across diverse environments. As a result, accessions with abiotic stress-resistance were identified that could be introgressed into commercial landscape trees (9).

Curators and CGCs recruited crop-specific experts to screen collections and core subsets for high-priority agronomic or horticultural traits. During this period, the NPGS funded about 150 germplasm evaluation projects that generated evaluation data for incorporation into GRIN. Curators and CGCs not only recorded critical germplasm evaluation data, but they also served as effective conduits for transferring information to customers and stakeholders.

The preceding users include public and private sector researchers and plant breeders who exploited the preceding information to select germplasm for further evaluation and incorporation into their research and breeding programs. Through such evaluations, a Mexican wild potato species distantly related to cultivated potato recently was discovered to be a major source of potato late blight resistance and genes from this species were transferred to cultivated potatoes (10). Most U.S. potato cultivars released by public-sector breeding programs during the last five years incorporated ARS-maintained and distributed germplasm selected on the basis of such evaluation data.

The new approaches developed for characterizing and evaluating NPGS genetic resources were often widely adopted by the research community. For example, methodologies developed for assessing NPGS vegetable germplasm were often utilized by vegetable seed companies as the standards for assessing breeding stocks and other germplasm. Also, novel non-destructive NIR analytical methods were developed to evaluate soybean, wheat, and sicklepod germplasm more effectively for oil, protein, and moisture content (11).

An innovative evaluation effort for wheat developed databases comprising information about host-plant resistance to pathogens. *Fusarium* head blight and scab of wheat are of paramount interest to wheat researchers, so a regional program for evaluating hard red spring and durum wheat germplasm was coordinated by ARS, and produced multilocation evaluations of hundreds of wheat lines for *Fusarium* head blight and scab resistance. The evaluation data were posted on the World Wide Web (12): ultimately, access to these data will contribute to reducing disease incidence in growers' fields.

Although many species maintained by the NPGS are represented by relatively few accessions, some collections of crop species are very large, making systematic evaluation and characterization enormous tasks. For example, the U.S. maintains more than 8,000 different types of peanuts collected from around the world. This collection includes many traits of potentially significant positive economic impact on commercial peanut production, but the limiting factors for identifying them are the time and expense needed to screen the entire collection for any particular trait. Consequently, a core subset of more than 800 accessions and a smaller subset of the latter (a "mini core subset of about 100 samples) was developed to improve the efficiency of identifying these valuable genes (13). Core subsets for apple, potatos, small grains and several other large NPGS collections were developed to improve the efficiency of germplasm evaluations. Through these evaluations, sources of resistance to economically significant pathogens were identified and many genes from otherwise unadapted germplasm was exploited by U.S. breeders. For example, weevil resistance identified in unadapted pea germplasm provided a wider suite of resistance genes for pea breeding programs.

Evaluations of genetic diversity in wild insect and microbe populations have also yielded important results. For example, diverse forms of fungal endophytes identified in NPGS grass collections furnished new sources of insect resistance for research (14). DNA marker analyses of bacteria uncovered a very sharp genetic boundary between different northern corn rootworm (NCR) populations. Distinct strains of the bacterium *Wolbachia* infested NCR on either side of the genetic boundary, suggesting that the bacterium is likely a primary cause of the NCR population divergence. The strains of the bacterium inhibit NCR reproduction between individual insects with different bacterial strains and resulted in three distinct NCR genetic populations: two populations with mutually incompatible *Wolbachia* and a third population that is uninfected (15). This finding has significant implications for developing IPM programs for this important maize pest.

> **Impact:** Germplasm characterization and evaluation helped breeders and geneticists identify novel genetic variation valuable for crop improvement. Availability of new characterization data enabled germplasm users to better predict what germplasm will be most useful to them, and how to use it more efficiently. The development of improved germplasm evaluation techniques and core subsets for large germplasm collections encouraged more frequent germplasm utilization. A byproduct of these evaluations was the identification of duplicate and mixed accessions, as well as gaps in collections (see Problem Area 1c). Another benefit of evaluations was the identification of patterns of genetic diversity potentially valuable for predicting heterotic responses, crossing incompatibilities, and other genetic properties important for optimizing plant breeding progress.

Problem Area Ie - Technology Transfer of Genetic Resources and Associated Information

Outcomes, Goals, and Commitments: The U.S. national germplasm system differs from many other national and international genebanks by its clear, long-established commitment to effectively transfer massive volumes of genetic resources and associated information to researchers, so as to catalyze the ultimate outcomes of crop improvement, economic development or improved environmental conditions. USDA/ARS genebanks are committed to transferring expeditiously healthy and vigorous genetic resources, accompanied by key descriptive information and documentation (e.g., handling/cultivation needs and methods, phytosanitary status). Information must be communicated promptly and effectively in response to queries.

USDA/ARS genebanks also seek to develop and implement new mechanisms for facilitating germplasm and information transfer. New diagnostic tests may be needed to assess pathogen/disease status and accelerate processing of germplasm through quarantine. In some cases, training in genetic resource management methods, database operations and use, and related activities must be conducted.

Selected Accomplishments: Germplasm transfer within the U.S., and especially internationally, is complicated by strict and complex quarantine requirements, transportation regulations, and documentation systems. Reconciling the complicated international system of germplasm transfer with the U.S. philosophy of open access to genebank material is especially problematic.

Despite the preceding challenges, NP 301 scientists transferred expeditiously enormous volumes of healthy, well-documented germplasm samples and associated information. The distribution numbers are impressive. The NPGS annually distributed an average of more than 140,000 samples, which constitutes about 30% of the entire collection. During the last five years, the number of individual requests for NPGS samples increased by more than 16%, and currently is more than 4,500 requests per year.

For example, the NPGS genebank in Aberdeen, ID distributed more than 130,000 seed samples of wheat, oats, barley, and rice in response to about 2,500 separate requests during the past five years. Quality and other important evaluation data for core subsets were also distributed (1). Similarly, the NPGS genebank in Ames, Iowa distributed more than 44,000 samples of maize, oilseeds, vegetables, and specialty crops to fulfill more than 2,500 requests. Approximately 20-25% of the Ames collection was distributed each year, with about 20% shipped internationally. Likewise, the crop genebank in Griffin, Georgia distributed more than 170,000 accessions to fulfill more than 3,200 orders, with 88% distributed U.S. requesters and 12% internationally.

To help ensure that healthy genetic resources were distributed, descriptions and diagnostic tests for pathogens and pests that infect plant propagules were developed and information about them were recorded in GRIN and other public databases so as to better inform clientele about phytosanitary issues. For example, a website (2) was developed to describe fungi infecting legumes, including Asian soybean rust and other rust fungi that might be confused with the former. Each fungus was described and interactive keys were constructed that enabled rapid identification of rust fungi, powdery and downy mildews, scab fungi, and other invasive fungi. A similar system was developed for identifying both target fungi and candidate strains of *Trichoderma* for biological control of the preceding fungi.

Although transferring genetic resources and information to researchers and breeders are the ultimate goals for NP 301 Research Component 1, informing the general public about the intrinsic value of genetic resources is also important. To this end, during the last five years, such information was disseminated to the approximately 500,000 people who visited the ornamental plants collection at the U.S. National Arboretum annually, to the 5,000,000 annual "visitors" to the USNA web site, and to a newspaper readership of ca. 800,000 readers weekly.

> **Impact:** The impact of the preceding technology transfer on U.S. and global agricultural sustainability and research productivity is immense. For example, the Materials and Methods sections of numerous articles in recent issues of research journals such as *Crop Science* or *HortScience* cite germplasm, readily identified by a "PI number," that originated in the NPGS. Many of the genetic resources distributed for research and breeding by USDA/ARS genebanks were either no longer available elsewhere or no longer accessible without restriction due to rapidly changing global germplasm exchange policies. This broad genetic diversity enabled research programs to efficiently produce new cultivars, develop new knowledge, discover value-added uses, and preserve U.S. food security. Furthermore, ready access to detailed scientific descriptions of pathogens and pests provided invaluable tools for Federal and State agencies that protect U.S. borders from invasive species. This National Program's unparalleled record of genetic

resource and information transfer served as an important factor for successfully negotiating access to germplasm internationally. Foreign governments and institutions sometimes granted such access because of the U.S.'s long record of permitting unrestricted access to its genebank materials.

RESEARCH COMPONENT II: GENOME CHARACTERIZATION AND GENETIC IMPROVEMENT

The genetic resources managed by USDA/ARS genebanks (see Research Component 1) are the basis for improving the productivity, quality, and product values of crops, and for capitalizing on the use of microbes and beneficial insects to fulfill the needs of U.S. agriculture. Characterization of crop, microbe, and insect genomes provide a mechanism for facilitating, and often enabling, significant technological advances that add value to genetic resources and result in elite varieties for commercial production. This Research Component of the NP 301 Action Plan is committed to achieving the goals of identifying agriculturally-valuable genes, and exploiting them via genetic improvement projects.

Problem Area IIa - Genome Characterization

Outcomes, Goals, and Commitments: Five years ago, USDA/ARS committed to conducting an extensive program of plant, microbial, and insect genome characterization, focusing particularly on traits that can be exploited for genetic improvement. Genome characterization provides a foundation for developing and applying new strategies for accelerated genetic improvement via manipulating both simply-inherited traits and quantitative traits.

Problem Area IIa constitutes much of National Program 301. It was feasible to report only a sampling of this Problem Area's complete spectrum of accomplishments. Those achievements described here were expected to yield economic advantages and contribute to a more stable supply of high quality agricultural crop products for customer/stakeholders, while reducing agricultural impacts upon the environment. These selected accomplishment and impact statements were reported under eight general thematic categories: *(1) tools for genetic/genomic analyses, (2) special research populations and/or genetic stocks, (3) genetic determinants of important traits, (4) genetic marker systems, (5) genomic characterization and analysis techniques using model species, (6) mapping agriculturally-important genes, (7) genetic/genomic databases using model species, and (8) advances in genetic/genomic theory.*

(1) *Tools for Genetic/Genomic Analyses*

Selected Accomplishments: NP 301 scientists and university partners have made a major contribution to developing medium and high density genetic maps, incorporating many phenotypic traits and molecular markers, for numerous crops. These include all major species grown on very large scale, as well as many specialty crops with low production acreage but high value to growers. Markers requiring DNA sequence information, such as SSRs, SNPs, and candidate gene markers were also developed for such diverse crops as wheat (1), sorghum, soybean, peanut, cotton, alfalfa, buffelgrass, citrus, strawberry (2), blueberry, cacao, hydrangea and crape myrtle. In many cases these markers were used to construct molecular genetic linkage

maps in crop species such as wheat, sorghum, cotton, soybean (3), alfalfa, buffelgrass and cacao. These same markers were applied to germplasm characterization as a tool for genetic resource management as described in Research Component I.

In certain instances, ARS scientists took advantage of the wealth of knowledge available in better-characterized species to develop tools for less well-characterized crops. For example, the syntenic relationship between rye and rice genomes enabled rice sequence to be applied to designing PCR-based DNA markers in a region of rye-rice synteny. In this region the rye genome contained an important gene for resistance to high levels of soil aluminum. The resulting rice-derived DNA markers have proven useful for identifying progeny from rye x wheat hybrids that carry the gene for aluminum tolerance (4). Similarly, the availability of rice genome sequences made it possible to discover the causal SNP in the *Pi-ta* rice blast resistance gene. This DNA polymorphism, and others in close proximity, could then be applied to select for rice blast resistance (5).

ARS scientists constructed BAC libraries in order to develop SSR markers in cotton and cacao, for map-based cloning in tomato, and fluorescent in situ hybridization in potato. In the case of sorghum, an integrated physical and genetic map was aligned with the draft genome sequence of rice, thereby serving as a valuable resource for comparative analysis of grass genomes (6). In soybean, BAC clones constituted key "infrastructure" for a physical map, by anchoring a large number of BAC contigs to the genetic map.

Structural genomics of insects was an important component of the ARS research portfolio over the past five years. ARS scientists formed part of the leadership of the collaborative Honey Bee Genome Project that generated an annotated draft genome sequence of the honeybee. From this draft sequence more than 2,000 polymorphic markers were identified and are being used for breeding and for disease-gene identification. Similarly, a high-resolution genetic map was constructed for the genome of the red flour beetle, *Tribolium castaneum*, an important pest of grain products and a facile genetic model species for laboratory research. The map consists of more than 400 genes, expressed sequences, and genomic BAC clones covering all ten chromosomes (7). In addition, a SSR-based map of Hessian fly, an important pest on small grains, is being constructed and integrated into the existing genetic linkage map.

Impact: Genetic markers and genetic and physical maps are vital prerequisites for a broad spectrum of genomic research, from gene/QTL discovery to comparative genomics to whole genome sequencing. They provide basic biological information of general interest to science, as well as tools applicable to managing genetic resources and developing crop cultivars. The leading roles played by ARS scientists in developing genetic markers in wheat, sorghum, soybean, peanut, cotton, alfalfa, forage grasses, cacao, blueberry, strawberry, citrus, hydrangea, crape myrtle and other species were and continue to be instrumental in improving the density and utility of genome maps. NP 301 research advanced international cooperative efforts to develop genetic maps for sugar cane, cacao, cotton, and bananas. Genetic markers, precisely targeted to genes controlling biotic and abiotic stress in exotic germplasm, now enable those genes to be introgressed into stress tolerant adapted germplasm and crop cultivars. Anchoring of physical to genetic maps in crops such as sorghum and soybean

enabled comparative genome analysis and provided the frameworks for targeted and whole genome sequence analysis (8).

The availability of a public BAC library in soybean and other crops catalyzed several initiatives to expand the scope of genomics research. The BAC clones constituted the key component of a physical map that enables the translation of genomic information from model plants to crops and is setting the stage for whole genome sequencing projects. When completed, this will generate a wealth of information for developing genetic markers for breeding and for understanding the genome structure and organization. This is being realized, for example, in soybean where ARS scientists and their collaborators are completing a physical map anchored to the genetic map.

As in crop genomics research, NP 301 and other ARS scientists engaged in insect genomics research made significant contributions to research community building over the past five years. The impact of the newly released annotated draft sequence of the Honey Bee Genome is only beginning to be recognized. The complete genome sequence provides easy access to PCR-based genetic markers and lists of candidate genes for disease responsiveness and other traits of importance in honeybee improvement (9). Continued refinement of the genetic map of Hessian fly now enables research on the evolution of virulence to new resistance genes being deployed in wheat cultivars. Similarly, the complete sequence of the red flour beetle furnishes a ready source of genetic markers, as well as candidate biopesticide genes, which may enable novel control measures to be developed for this insect pest. Furthermore, the red flour beetle whole genome sequence is the first among the coleopterans, the largest group of insects that cause billions of dollars in crop losses. Thus, availability of the red flour beetle whole genome sequence has potential implications for the controlling other insect pests.

(2) ***Special Research Populations and/or Genetic Stocks***

Selected Accomplishments: NP 301 scientists produced special research populations and genetic stocks in a wide range of crops and their relatives. A typical example of this type of work is the 20 hexaploid wheat populations developed, via marker assisted backcrossing, to incorporate resistance to Barley Yellow Dwarf Virus, strawbreaker foot rot, stripe rust, preharvest sprouting and Hessian fly. Such development of backcross populations provides important germplasm to U.S. wheat breeders and the genetic markers that are identified can be used to introgress these resistances into a range of wheat market classes.

Another example of a set of unique germplasm developed by ARS scientists and their collaborators was oat lines, each carrying a different segment or segments of a specific maize chromosome (10). These materials were distributed to more than 30 public and industry laboratories around the world as highly important genetic tools for analyzing the maize genome and also as possible sources of non-genetically-engineered oat germplasm containing genes from maize.

Research to broaden the genetic base of the U.S. maize was conducted to ensure continued genetic gains in productivity. ARS scientists crossed U.S. inbreds with exotic tropical accessions

to produce progeny that, as estimated by SSR marker characterization, derived 33% of their alleles from the tropical source (11).

In the case of cotton, a newly defined standard panel of cotton genotypes developed by ARS scientists is now widely used by cotton geneticists/genomicists to systematically evaluate the utility of new genetic markers. This panel of 12 diverse genotypes was identified and established as the standard set of lines for molecular marker characterization. All potential genetic markers are being screened for this panel of genotypes by U.S. cotton workers (12). These evaluations minimize marker redundancy and provide an assessment of potential suitability for genetic mapping and for breeding programs. Similarly, in less well-characterized species, such as *Hydrangea* and crape myrtle, ARS scientists have established large mapping populations for constructing the first molecular genetic linkage maps of these species.

When naturally occurring genetic variation is unavailable, mutagenesis can sometimes create genetic variants with required characteristics. For example, in many grain crops, phosphorus is bound in phytic acid making that element biologically unavailable to non-ruminants. No naturally occurring variation is available for this trait. Low phytate mutants of barley, wheat, rice and soybean were created via mutagenesis, and molecular markers tightly linked to the low phytic acid loci were identified. These mutants represent an important advance in grain nutritional quality (14) and may have a potential impact on phosphorous runoff from animal production enterprises. Molecular markers tightly linked to low phytic acid traits were identified, which will expedite introgressing the traits into adapted cultivars.

In addition, gene mutations sometimes enable a better understanding of the genetic and physiological bases for expression of a given phenotype. Lesion mimic mutants with enhanced disease resistance and susceptible mutants helped reveal the mechanism of disease resistance and the function of the important *Pi-ta* rice blast resistance gene (13).

> **Impact:** Developing special populations and genetic stocks is one route by which ARS scientists deliver research tools that can lead directly to new cultivars for U.S. producers or to a better understanding of the genetics and/or physiology of plant, insect and microbial genomes. Special populations and genetic stocks are broadly shared through distribution by individual NP 301 scientists, as well as by ARS genebanks, especially Genetic Stock Centers (Maize Genetic Stock Center and Rice Genetic Stock Center). Germplasm with specific traits, e.g., low phytate, are immediately useful in cultivar development, whereas other mutants, such as the lesion mimic mutants in rice, will serve as tools for elucidating mechanisms of host-plant resistance. ARS scientists distributed these mutants to researchers studying programmed cell death, the hypersensitive reaction to avirulent pathogens and the general process of disease defense response. Such studies should lead to a better understanding of the mechanisms of host-plant disease response and the development of more durable and stable resistance.

(3) *Genetic Determinants of Important Traits*

Selected Accomplishments: ARS scientists and their collaborators conducted research on a wide range of wheat traits to identify the genome position of unique genes or QTL and to

develop genetic markers to permit their efficient introgression into adapted cultivars. Using tetraploid wheat accessions and synthetic hexaploid wheats, ARS scientists evaluated reaction to Fusarium Head Blight (FHB), tan spot, and *Stagonospora nodorum* blotch, and identified a number of novel sources of resistance to these diseases. Molecular markers were used to determine the chromosomal locations of FHB resistance QTLs derived from *Triticum dicoccoides* in tetraploid mapping populations. The ARS-managed U.S. Wheat and Barley Scab Initiative has coordinated and supported identification of QTLs for FHB resistance in all market classes of wheat and barley for use in genotyping and genetic improvement. The chromosomal locations of QTL for race-specific resistance to *Stagonospora nodorum* blotch and race-nonspecific resistance to tan spot were defined and were mapped (15).

Novel genes were identified that are involved in response to Hessian fly damage. These included several lectin genes (16), as well as an up-regulated flavanone-3-hydroxylase gene, which may contribute to the production of toxic tannins or reactive oxygen species that damage larval tissues. Two new wheat genes conferring resistance to the Hessian fly were identified, mapped, and molecular markers developed. Similar research aimed at the QTL controlling biotic stress resistance in barley was undertaken to discover and expedite the incorporation of QTL into agronomically superior cultivars. These included QTLs for morphological traits, FHB resistance, low DON (deoxynivalenol or vomitoxin, associated with FHB), and crown rust resistance (17-19).

In addition, an efficient and effective molecular marker technique, target region amplified polymorphism (TRAP), was developed that takes advantage of the more than 60,000 EST sequences available for sunflower. The TRAP marker technique was used to identify a DNA marker associated with resistance to all known downy mildew races of sunflower (20).

QTLs for resistance to high temperature stress in wheat and cold seedling vigor in sorghum were successfully identified. Cold-responsive genes encoding antioxidants and phospholipid synthesis and modification systems were characterized and quantified in wheat. Phospholipid structures were found to be modified in response to cold and evidence of phospholipid involvement in signal transduction was found (21,25).

Traits related to the mode of plant reproduction were evaluated and DNA markers identified in the buffelgrass linkage map in close proximity to the gene(s) controlling apomixis (asexual reproduction through seed) (22). In addition, two markers linked to dioecy (separate male and female plants) were identified on the Texas bluegrass map (23). These DNA markers can be used to facilitate genetic improvement of these traits in two important forage grass species.

QTL discovery and marker assisted selection have also progressed in horticultural species. In cacao, two QTL, one accounting for 40% of the variation in resistance to the witches' broom disease pathogen were identified (24) and promise to have a beneficial impact on this perennial species. In addition, QTL discovery and marker assisted selection for disease resistance in *Hydrangea*, strawberry and blueberry have been initiated.

Impact: The preceding results will advance the development of enhanced germplasm or cultivars that provide economic advantage to the producer. Resistant germplasm, as well as

the associated genetic markers, advance introgression of disease resistance and weather stress tolerance into adapted cultivars.

(4) ***Genetic Marker Systems***

Selected Accomplishments: The frequency of SNPs in soybean germplasm was determined via the sequence analysis of fragments of more than 100 genes in 25 diverse cultivars and other genotypes. Sequence diversity in soybean genes was determined to be 1/10 that of maize. A set of six maximally diverse soybean genotypes was identified from the 25 genotypes to optimize SNP discovery in soybean genes (26). SNP discovery in 4,000 unigenes, via the re-sequencing of the six genotypes, detected more than 1,500 SNP-containing unigenes. ARS scientists and their collaborators are genetically mapping these SNPs and integrating them into the existing SSR and RFLP-based framework genetic linkage map.

Impact: SNP DNA markers promise to be a tool for the creation of high density genetic maps and effective throughput genotyping useful in genetic resource management and plant breeding. Research on the frequency of SNPs in soybean provided the first estimate of nucleotide diversity (SNP frequency) in soybean genes and defined an approach for large scale SNP discovery in soybean genes. The discovery and genetic mapping of more than 1,500 unigenes is the first step towards developing a SNP-based genetic linkage map. It will provide soybean researchers with a tool for high throughput genetic analysis to identify genes that govern agriculturally important traits, and for developing marker-assisted selection methods that accelerate soybean improvement.

(5) ***Genomic Characterization and Analysis Techniques***

Selected Accomplishments: ARS scientists cloned the *Arabidopsis Clavata3* (*CLV*3) gene and determined the mechanism by which *CLV*3 functions to control stem cell accumulation in shoot and floral meristems. The transcription of a number of *CLE* (*CLV*-related) genes was determined to be highly tissue specific and stage-specific indicating that they may function as extracellular signaling molecules in diverse pathways during plant growth and development (27).

Similarly, the Sleepy1 *(SLY*1*)* gene in *Arabidopsis* was cloned by ARS scientists (28). It was determined that *SLY*1 transmits the GA (giberellic acid) signal by targeting the DELLA proteins for degradation. DELLA proteins are thought to act as repressors of GA-regulated processes. *SLY*1 encodes a subunit of ubiquitin that binds to DELLA proteins and targets them for degradation. The cloning of *SLY1* is the basis for a new model for GA signaling.

Other ARS research that relied on a model system had the objective of developing an understanding of the regulation of pollen development and pollen-pistil interactions. ESTs from embryo sac, sperm cells and egg cells were characterized in order to identify promoter elements that direct gamete-specific gene expression. This work identified molecules that mediate signaling during pollen tube growth and a protein in the female that enhances pollen tube growth (29, 30). By fully understanding pollen-pistil interactions, as well as egg sperm interactions, genetic engineering may enable the successful hybridization of distantly related crop species.

Impact: The impact of translational genomics, that is, the application of genomic information from relatively well characterized model species to species for which few genomic resources exist, is yet to be widely realized. However, ARS scientists and other researchers have initiated work in model species that promises to positively impact crop plants. The *Arabidopsis CLV*3 gene, which was identified as the first small polypeptide ligand known to regulate plant development, provides an example since this research uncovered a family of related molecules that are conserved among higher plants. From this research a model was formulated describing how *CLV*3 acts as a diffusible signaling molecule that communicates cell fate information to neighboring cells. This model serves as a new paradigm for ligand-receptor signal transduction in plant stem cell maintenance that has been widely adopted by the plant biology community (31). Likewise, the cloning and characterization of the *SLY*1 gene was a fundamental advance in the understanding of GA signaling. The cloning of *SLY*1 by ARS scientists and the cloning of its rice homolog *GID*2 by Makoto Matsuoka, Japan, were the first reports indicating that the GA signal is transmitted via proteolysis. Mutations in the GA-related DELLA genes provided the semi-dwarf crop varieties that were the basis of the Green Revolution. Research shows that GA regulates the DELLA/Green Revolution genes via SLY1/GID2, and opens up new avenues for GA research world-wide on the role of the ubiquitin-proteasome pathway in GA signaling (32).

(6) ***Mapping Agriculturally-Important Genes***

Selected Accomplishments: A number of efforts aimed at the cloning of agriculturally important genes were completed and are on-going in NP301, as well as National Program 302, Plant Biological and Molecular Processes. In one such project, the pollen fertility restorer gene, *Rf1*, from sorghum was cloned and sequenced and its cloning facilitated the development of highly accurate genetic markers for germplasm characterization (33). Another project resulted in the isolation and characterization of the tomato *RIPENING-INHIBITOR* (*RIN*) gene that represents a key regulator of fruit ripening (34). It was demonstrated that the *RIN* gene is a member of the MADS-box family of transcriptional regulators and is highly conserved among diverse crop species producing fleshy fruit (e.g. tomato, pepper, strawberry, and banana).

Impact: The cloning of agriculturally important genes is a difficult undertaking in most crop species in which genetic and physical maps are often rudimentary. Nonetheless, the sorghum pollen fertility restorer gene, *Rf1*, was successfully cloned and represents the first agriculturally-important gene to be positionally cloned from sorghum. In addition, ARS scientists lead collaborative efforts to develop and apply bioinformatic software for positioning genes on genetic maps (gene ontology) in other crops species. For example, statistical modeling of redundant BAC `hits' using mapped genetic markers indicated that most soybean genes will be found in approximately 25% of the genome. This knowledge has improved the strategy for genome sequencing in soybean and has saved many millions of dollars in sequencing costs.

(7) ***Expansion of Genetic/Genomic Database Resources using Model Species***

Selected Accomplishments: ARS has established and supports the Germplasm Resources Information Network (GRIN, see Research Component I); SoyBase, the soybean genome database; GrainGenes, the Triticeae and related species genome database; Gramene, A Comparative Mapping Resource for Grains, MaizeGDB (Maize Genetics and Genomics Database), as well as a number of other crop specific databases (described further in Research Component III). NP 301 and other ARS scientists have deposited their genomic characterization data in these databases, to promote public access and use, particularly by crop geneticist and breeders. Submissions range from individual genes to whole genome draft sequences. Individual genes include submission of the mRNA sequence of the *Arabidopsis* F-box family protein/*SLEEPY1* to GenBank, full-length *Arabidopsis CLE* (*CLV*3-like) sequences to GenBank and TAIR (The Arabidopsis Information Resource) and the sugarbeet sucrose synthase mRNA to GenBank (35).

Extensive EST sequences from a number of species including soybean, peanut, sorghum, wheat, barley, sugarbeet, buffelgrass, blueberry and honey bee have been deposited by ARS scientists and collaborators in GenBank and other species-specific genome databases. End sequences of soybean BAC clones were submitted to GenBank and information regarding the presence of SNPs in soybean genes was submitted to dbSNP at NCBI. SSR marker information for cotton was made available in GenBank and CottonDB. Similar SSR data for peanut were submitted to GenBank and wheat SSR information was submitted to GrainGenes. Likewise, genetic maps of a number of species including wheat, sorghum and soybean have been significantly enhanced in the past five years by ARS scientists. An extensive SSR allele size database with data for 236 rice accessions is accessible (36). A similar allele size database of 380 accessions of the Citrus Variety Collection genotyped with 24 SSR was made public by ARS scientists (37). Extensive information relating to genetic markers, alignment of the sorghum genetic map with the draft sequence of rice, as well as other genomic data are available in Gramene.

In the area of insect genomics, ARS scientists and their collaborators working in red flour beetle and honeybee genomics have created specialty databases. Beetlebase and BeeBase both contain extensive compilations of genomics data for their respective species.

> **Impact**: The research community has benefited from ready access to this new information. Importantly, the data deposition and tool development have strengthened research community building for full genome sequencing and genetic improvement.

(8) ***Advances of Genetic/Genomic Theory***

Selected Accomplishments: ARS scientists developed new statistical approaches to identify candidate genes in maize more precisely and with more experimental control regardless of population structure. This candidate gene association approach can increase resolution up to 2,000 fold versus standard QTL mapping and can be applied to any crop species (38). Similarly, ARS rice researchers and collaborators have analyzed the population structure of a group of more than 230 diverse rice accessions (39). These data were collected as the starting point for the application of genetic association analysis or association mapping for the discovery of genes/QTL using pre-existing germplasm without the need to develop mapping populations. The association mapping approach was successfully applied by ARS scientists working in cacao.

SSR markers were used to detect associations between productivity QTL and alleles at SSR loci (40).

While transposon tagging has been available in maize, and *Arabidopsis* and other plant species for some time, such a system does not exist in many important crop plants. The occurrence of homologs of maize transposons in wheat was determined by Southern hybridization and by searching wheat EST sequences. Homologs of the *Ac/Ds* transposon were less well represented in the wheat genome than either *mutator* or *En/Spm*. Thus, the *Ac/Ds* transposon appears to be an excellent candidate for a wheat tagging system and its development is being pursued (41).

Radiation hybrids have greatly facilitated the development of genetic maps in mammalian and other vertebrate species. While this type of system is not available in plants, ARS scientists with their collaborators have begun to investigate analogous approaches. Ten self-fertile oat lines, each with a different complete maize chromosome were used as the starting point for research. The added maize chromosome in these plants was broken into smaller segments by treatment with gamma radiation. Using these procedures novel sets of plants, each carrying a different segment or segments of a corn chromosome, have been produced from each of the original ten plants. The resulting oat lines are being characterized with maize-derived molecular markers to determine the presence and extent of intact maize DNA (42). This genetic mapping system in plants obviates the need to create mapping populations and dependence on genetic polymorphism.

Determinate versus indeterminate growth habit and senescence are processes that are tied to timely marketing of crop products. ARS scientists discovered a gene family in maize (KNOX genes) that play a role in maintaining an indeterminate state that can prevent senescence (43). Other scientists demonstrated that the same gene family can control senescence in horticultural crops. In addition, the same gene family regulates lignin biosynthesis and plays a key role in plant development. The discovery of key genes controlling plant development and the senescence process provide breeders with markers for meristematic activity and opens the door for producers to control when their crop is ready for market. This has the potential to save many millions of dollars in lost produce due to over ripening, or senescence before optimal crop production.

Few details are known about how sex chromosomes are involved in plant sex determination even though proper expression of sex is the single most important contributor to yield of all fruit and seed producing crops. ARS scientists and their collaborators discovered a primitive sex chromosome in papaya having a very small male-specific region of 4-5 Mb in size compared to the male-specific region that is 10-fold larger (60-70 Mb) in humans and 100-fold larger (500-600 Mb) in the most studied plant sex chromosome of white campion (44). The sex chromosome is postulated to resemble the ancestor of the human Y chromosome as it existed 240-320 million years ago so that sequencing this chromosome provides a unique opportunity to test hypotheses and theories about sex chromosome evolution. Fine mapping and sequencing of the sex determining region should uncover the sex determining gene that will suggest mechanisms responsible for evolution of sex chromosomes and be used to develop true breeding hermaphrodite lines needed by papaya producers.

How bacteria evolve is important in the study of plant symbionts as well as plant pathogens. ARS scientists developed statistical evidence that different genes provide dissimilar evolutionary signals in different bacteria and that this is due to lateral transfer and recombination of portions of the genes among distantly related bacteria (45). The fact that lateral transfer of genetic material can occur between distantly related bacteria may have a major impact on the study of bacterial evolution.

Impact: Plant geneticists have traditionally relied on the inheritance or segregation patterns of phenotypic traits to make progressive gains in crop productivity and quality. Research progress and accomplishments of National Program 301 are manifested in major advances in population genetics that will enable more knowledgeable and effective decisions in future crop improvement. For example, the application of genetic association or 'linkage disequilibrium' theory by ARS geneticists has enabled a better understanding of the impact of natural and artificial selection on the loss of genetic diversity in modern maize and soybean genomes (46). Whereas estimates up to 50% of the genetic diversity resident in wild accessions of these crops may have been lost during the transition to land races, research shows that artificial selection (breeding) has affected only a limited number of genes during development of elite cultivars. This knowledge demonstrates that exotic or wild species are a substantial genetic resource of 'untapped' genes for enhancement of modern cultivars, and also helps identify the genes that serve as the basis for crop improvement in domesticated crops. Thus, successful application of genetic association analysis will accelerate the rate of gene/QTL discovery in plants, and improve the ability of geneticists to "mine" for useful genes for crop enhancement.

Problem Area IIb – Genetic Improvement

Outcomes, Goals, and Commitments: The goal of this problem area is to provide U.S. agriculture with superior genetic resources to ensure an ample and high quality supply of food and feeds. The expectation is that superior cultivars and genetically-enhanced populations will result from introduction of untapped sources of genetic diversity into breeding populations, and the introgression of novel high value traits into standard breeding lines and genepools, for beneficial microbes and insects as well as plants.

National Program 301 scientists, often in partnership with universities and industry, have committed to strengthening significantly U.S. capacity for developing advanced breeding lines and promising new varieties. Because Problem Area IIb accounts for a large portion of National Program 301, accomplishments and impact statements are reported under six categories: *(1) release of superior genetic resources, (2) capitalizing on untapped genetic diversity, (3) genetic mitigation of biotic and abiotic stress, (4) genetic improvement of product quality and value, (5) new genetic methods for crop enhancement, and (6) statistical approaches to optimize breeding progress*.

The following statements represent a sampling of the National Program 301 accomplishments that are expected to yield significant advances toward improving disease resistance, tolerance to abiotic stresses and end-product quality in varieties and germplasm for U.S. agriculture.

(1) Release of Superior Genetic Resources

Selected Accomplishments: NP301 scientists take the lead in developing and evaluating superior varieties, breeding lines, and enhanced germplasm for numerous other agronomic and horticultural crops. Selected examples follow and more information is available in NP301 Annual Reports at: http://www.ars.usda.gov/research/programs/programs.htm?NP_CODE=301):

The new aphid-resistance barley cultivar (3) developed by ARS will reduce the need for pesticide application and stabilize production. In certain areas of the intermountain West, barley production has been abandoned because of recurring Russian wheat aphid infestations or required insecticide applications. The new ARS cultivar will provide producers in dryland, low-productivity environments with the option of growing barley again (3).

The potato variety, 'Defender,' developed and released in 2004, is the first late blight resistant potato variety suitable for processing into potato products (4). Defender will dramatically reduce and in certain growing environments, eliminate fungicide applications for the control of late blight. Late blight is the single most important potato foliar and tuber disease worldwide, costing $100 million in the United States alone to control on an annual basis. This cultivar will improve farm profitability by reducing fungicide applications, which may save farmers up to $150 per acre, and by reducing fungicide impact on the environment.

Three spring dry pea varieties were released with increased yield and end-use quality characteristics and a chickpea (Sierra) with resistance to ascochyta blight. Sierra is projected to become the predominant chickpea variety in the Pacific Northwest over the next several years.

ARS soft winter white and club wheat varieties with enhanced disease resistance and end-product quality have been grown on more than 1.5 million acres in the Pacific Northwest each year for the past five years. Improved germplasm from the ARS hard winter and spring wheat program (Kansas) was utilized in the development of commercial wheat varieties, Thunderbolt and Overley. The combined acreage of Thunderbolt and Overley in Kansas in the 2004/2005 season was 400,000 acres and it is expected to increase significantly next year. Additional acres of these two varieties are planted in Oklahoma, Texas, Colorado, and Nebraska. Since 2000, the ARS wheat germplasm program in Kansas has received 261 requests for seed of their germplasm lines by wheat breeders and geneticists, attesting to the impact on wheat breeding programs worldwide.

ARS scientists in cooperation with university collaborators released two peanut cultivars that have enhanced resistance to Sclerotinia blight and high oleic acid content. *Sclerotinia* blight is a fungal disease that severely limits peanut production in the southwestern United States. Increasing the level of oleic acid in peanut oil is a health benefit and improves the frying value of peanut oil. These lines have the potential to enhance the cash flow to peanut growers in Oklahoma and Texas by $6 million annually.

Sugarcane cultivars developed by ARS occupy more than 92% of the sugarcane production acreage in Florida and produce sugar worth more than $600 million annually (5). Similarly, new small fruit cultivars have been accepted by the commercial small fruit industry in the Pacific

Northwest as evidenced by the sale of over one million plants of new ARS strawberry cultivars in 2004 (6,7); high demand for two new ARS blackberry cultivars resulted in the "selling out" of nursery stock in 2005; the number one sales rank of an ARS blackberry cultivar in 2004, with 87% greater sales than the industry standard (8), and the number two sales rank of an ARS red raspberry cultivar.

ARS scientists throughout the South and Southwest are developing and releasing improved cotton varieties (9). Breeders have developed advanced lines with 10% higher fiber bundle strength and 23% lower short fiber content. This germplasm provides traits that will better support exports to the rapidly growing Chinese textile industry, now the largest buyer of U.S. cotton.

New ARS cultivars impact not only growers, but consumers as well. For example, thousands of trees of new varieties of plum and plumcot have been planted, and interest is high in further plantings. These new varieties provide both large and small growers with viable and more profitable varieties, enhancing the fruit array available in the market to consumers. ARS also released three Dutch elm disease-tolerant American elm varieties, which should help to bring back one of the most popular landscape trees to American cities and towns (10).

In addition to crop plant cultivar and germplasm releases, ARS has an active genetic improvement program in honeybee. Parasitic mites have presented a growing threat to the honey industry in recent years. ARS scientists have now identified honey bees that provide a source of genetic resistance to two of the major mite pests (*V. destructor* and *A. woodi*) (11,12). Queens carrying this resistance trait are being provided to commercial providers and are being used in commercial colonies (13). Beekeepers now have a genetic stock of honeybees that will ensure continued availability of resistant colonies for pollination and honey production. Selection and incorporation of the resistance genetics into colonies has been shown to produce good resistance to Varroa, excellent resistance to tracheal mites, while retaining quality honey production.

Impact: In the past five years, ARS researchers have made substantial progress in developing and releasing superior germplasms and cultivars across a diverse array of crops. The released cultivars have improved agronomic performance, horticultural performance, disease resistances, or value-added traits compared to other currently available varieties. ARS researchers have released new cultivars of at least 39 different plant species, including cereals, vegetables, legumes, fruits, nuts, forage or range grasses, and ornamentals (1). Many of these varieties have been rapidly accepted by growers, and several of these varieties will permit the expansion of crop acreages to regions that were previously not amenable to production of those crops, due to disease pressures. ARS breeding programs in some major crops, such as wheat and soybean, released commercially competitive cultivars that serve geographic regions or market types not targeted by private industry.

In addition to released cultivars, ARS scientists have developed new breeding lines or enhanced germplasm (2). This new germplasm has strengthened industry efforts, by incorporating unique genes for disease resistance, crop quality, and productivity from exotic germplasm sources and wild species relatives. The-improved germplasm can be used by state and private industry breeding programs to enhance their elite crop gene pools. For

many horticultural, forage, and minor agronomic crops, few other breeding programs exist to develop cultivars. For example, nearly all of the hybrid carrot and onion, and about half the cucumbers, grown in the United States are produced using USDA germplasm. Therefore, ARS cultivar development in these crops often has substantial and direct impact on food, feed, and fiber production in the United States.

(2) Capitalizing on Untapped Genetic Diversity in Crop Improvement

***Selected Accomplishments*:** Elite varieties and germplasm developed by ARS scientists often contribute to a broadening of crop genetic diversity. Those accomplishments are facilitated by National Program 301 research that has pioneered the identification of distinct 'gene pools' within the wild ancestors of modern crops, and has demonstrated the level of genetic gain achieved by the introgression of exotic germplasm into domestic genepools.

For example, characterization of the origin, description and pedigree of all public soybean cultivars produced in China, Japan and the United Stats between 1923 and 1995 by ARS scientists has revealed three distinct gene pools for soybean among these geographic regions (22). This knowledge facilitated knowledgeable decisions on parental selection to maximize the incorporation of untapped genetic diversity in germplasm enhancement, and lead to the development of the soybean germplasm, LG00-3372, which recently exhibited the highest yielding ability in the Soybean Uniform Preliminary Test IIIB. More than 1,000 breeding lines with exotic pedigrees have been transferred to industry breeding programs.

Increased genetic diversity in corn also was realized by the release of 105 new inbred lines from the Germplasm Enhancement of Maize (GEM) program since 2001 (14,15). The GEM project is an ARS-led partnership with public-sector and private-industry researchers. The goal of the cooperative effort is to introgress elite corn lines with promising exotic sources and characterize the resulting hybrids. The genetic resources produced by this unique public-private cooperative program are then publicly released for general use to broaden the genetic base of U.S. maize (16).

Examples of broadened genetic diversity in small grains include five germplasm releases of wheat having disease resistances and bread-making quality genes from wild relatives (*Elymus trachycaulus*, *Triticum timopheevii*, *Aegilops speltoides*, *Ae. tauschii*, and *Haynaldia villosa*) (17) and the wide-spread production of the ARS-released soft winter white wheat cultivar "Madsen" with strawbreaker foot rot (eyespot) resistance from *Aegilops ventricosa*. Improved rice germplasm also has been developed from related wild relative and from indica rice from Asia identified as having high yields and disease resistances. Several of the indica germplasm releases have grain quality comparable to U.S. cultivars (18). An unadapted barley landrace was the source of resistance for the release of the first Russian wheat aphid-resistant barley (3).

In the solanaceous crops, genes that influence fruit carotenoid content were identified in an accession of the wild tomato relative *Lycopersicon cheesmanii* and introgressed into cultivated tomato to enhance fruit nutritive value (19). European germplasm was used in developing a late blight resistant potato cultivar, and nematode (20) and late blight resistant potato breeding lines were derived from hybridizations of cultivated potato with its wild relatives.

Newly released great northern bean germplasm lines are unique and genetically diverse in that rust resistance genes present in these lines were derived from the two gene pools of the common bean; the Andean and Middle American gene pools (21).

An analysis of forty years of data on U.S. short-staple cotton (*Gossypium hirsutum*) variety performance, both commercially and in yield test plots, provided guidance that reshaped the priorities of the cotton industry. ARS scientists showed that, beginning in the mid-1980s, cotton yields and quality traits had reached a plateau. More importantly, the yield potential (from test plots grown under best conditions) was also on a plateau, indicating for the first time a failure to achieve genetic progress with new varieties. As a result of this demonstration, the National Cotton Council made cotton genetics one of its highest priorities and formed task forces to identify opportunities to support cotton genetics. Cotton Incorporated restructured its programs to emphasize broadening the crop's genetic base and training new plant breeders.

ARS has been able to develop and is now releasing high-yielding new germplasm with large advances in heat tolerance, disease resistance, and fiber strength and length. With cooperators, ARS has now also released numerous monosomic substitution lines carrying extra-long staple (*G. barbadense*) chromosomes introgressed into *G. hirsutum* germplasm. This new germplasm provides alleles for extraordinary enhancement of fiber quality (24).

ARS cotton breeding and genetics research have long been the basis for the extra-long staple industry in the United States. More recently, ARS has released five new lines with superior fiber qualities and tolerance to abiotic stress (23). A high fiber strength line, also developed from *G. barbadense*, has been reported to have a level of resistance to *Fusarium* wilt. *G. barbadense* germplasm also was used in developing six earlier-maturing commercial lines more tolerant to abiotic stress, and having a broadened base for insect and boll rot resistance.

Wide species hybridizations were successfully achieved in sunflower and woody ornamental species of *Hydrangea* (25) and *Clethra*. These unique hybrids provide breeders with access to desirable genetic traits not previously available for their genetic enhancement.

Range and forage grass releases from the ARS including wheatgrass, wildrye, ricegrass, needlegrass, bromegrass, bottlebrush squirreltail, and bluegrass species represent a diversity of improved native and introduced range grasses that have had little if any previous breeding history (26).

> **Impact:** Many ARS germplasm enhancement programs have identified unique germplasm from unadapted plant genetic resources and subsequently have hybridized this material with cultivated breeding lines. This "pre-breeding" effort effectively places the desired traits of the exotic germplasm in a form more amenable for use by public and private breeding programs. Use of these genetic resources by breeding programs has demonstrated direct positive impact on U.S. agriculture.

(3) Genetic Mitigation of Abiotic and Biotic Stresses

Selected Accomplishments: Disease resistance is an important component of genetic enhancement. Examples of success in the identification of resistant germplasm include: nineteen accessions of rice with resistance to 'straighthead', a disease concern of southern U.S. rice producers (31); three breeding lines of romaine lettuce with resistance to lettuce dieback (32), a disease that can only be effectively controlled through host plant resistance; new sources of resistance to mycotoxin contamination in maize (33); Race 1 powdery mildew resistant watermelon germplasm (34), race 2 resistance screening is ongoing; TARS-PT03-1, a U.S. adapted, small seeded, pinto line with a novel source of resistance to the root rot complex of fungi in common bean (35); strawberry and blueberry germplasm with multiple disease resistances; and three pest- and highly Dutch Elm Disease-tolerant American elms.

The incorporation of disease and pest resistance genes benefits producers, consumers, and the environment by reducing the need for pesticides. Examples include wheat germplasm lines with resistances to leaf rust, powdery mildew, and Fusarium head blight (17), and sugarbeet breeding lines and populations with multiple disease and pest resistances (45). ARS and a coalition of universities have completed research on wheat and barley *Fusarium* head blight through the U.S. Wheat and Barley Scab Initiative (35a). Germplasm collections have been screened for new sources of resistance and genetic selections for scab resistance have been advanced.

ARS germplasm with unique resistances to pests, pathogens, and abiotic stesses for numerous crops are being used by public and private breeding programs for the genetic improvement of sugarbeet (root maggot resistance), wheat (multiple resistances), rice (multiple resistances), barley (Russian wheat aphid), cotton (multiple resistances), maize (multiple resistances), sunflower (broom rape resistance), potato (nematode resistance), *Hydrangea* and *Clethra* (cold hardiness), pinto bean (resistance to soil pathogenic fungi), great northern bean (rust resistance), lettuce (lettuce dieback resistance), strawberry and blueberry (multiple resistances), soybean (charcoal and *Phytopthora* rots), and watermelon (powdery mildew resistance) (3,27,28). This material provides growers with savings in pesticide use, thereby benefiting the environment and consumers as well.

ARS researchers have provided leadership for evaluating advanced public breeding lines for variety release. For example, the U.S. Regional Soybean Preliminary and the Uniform tests for the northern and southern region are coordinated by ARS scientists. Four ARS wheat geneticists coordinate the U.S. regional wheat variety trials that include: Western Regional Nurseries, Eastern and Southern Soft Red Nurseries, Hard Winter Wheat Nursery Program, and Hard Red Spring Nurseries. Four regional ARS Wheat Quality Laboratories also provide end-product quality assessment for the variety trials. Similarly, ARS researchers coordinate the Oat and Barley Performance Nurseries. Barley malting quality of all U.S. varieties has been measured by the ARS Barley Malting Quality Laboratory. Other evaluations have been conducted in multiple ARS nurseries that assess disease resistance and weather tolerance. Examples include Fusarium Head Blight and cereal rust nurseries (wheat, barley), and the Uniform Oat Winter Hardiness Nursery. During the past 5 years the vast majority of U.S. wheat, barley, and oat varieties were evaluated in these ARS coordinated nurseries prior to release. Rice milling and end-product quality of potential new varieties have also been evaluated.

ARS's coordinated assessment of disease resistance provides U.S. capacity to respond to new disease threats. For example, recent ARS greenhouse evaluation of U.S. winter and spring wheat varieties indicated that many U.S. varieties lack resistance to a new wheat stem rust race emerging in Eastern Africa. In response, ARS developed a cooperative agreement with CIMMYT to conduct resistance screening of U.S. wheat varieties in Kenya in the summer of 2005. Within a few weeks, the ARS regional wheat nursery coordinators contacted all U.S. wheat breeding programs (private and public), collected seeds of their advanced breeding lines and current varieties, and sent more than 500 accessions to Kenya for stem rust resistance testing in May 2005.

Examples of accomplishments in the identification of pest resistance include: new sources of resistance to greenbug biotype G within improved cultivars of barley (36), and new sources of resistance to European corn borer, western corn rootworm, and fall armyworm in maize (37,38).

Enhanced resistance to environmental stresses also is an objective of many breeding programs. ARS researchers identified three recent collections of wild sunflower as having drought stress (39), and Argentine bluegrass germplasm was combined with Texas bluegrass to produce a population with traits that enables survival and persistence in arid, warm to hot environments (40).

In soybean, established projects are focusing on diseases such as charcoal rot, sudden death syndrome and *Phytophthora* root and stem rot. ARS scientists developed a rating system to monitor the incidence of this disease, and have released the first high-yielding charcoal rot resistant soybean germplasm. Other ARS scientists have identified and characterized new biotypes of pathogens for root rot and sudden death syndrome. For example, 45 races of *Phytophthora sojae* are known, with 19 new races described in the last ten years (41). These investigations compare and describe frequencies and virulence of each race.

> **Impact:** ARS research on crop quality permits identification of superior germplasm and impacts cultivar releases. Impacts from germplasm identification are expected to be observed in the future, but historically, the identification of new sources of disease resistance is a critical first step toward the deployment of new resistances in adapted cultivars. For example, yield reductions due to *Phytophthora* root rot of soybean are estimated to be in the 125 million dollar range, annually. Identification and documentation of new races or biotypes of the pathogen is critical to planning disease control strategies. As a result of this research geneticists and breeders now have access to characterized isolates of this pathogen, thus opening the way to enhanced screening programs that will enhance germplasm and reduce yield loss to this disease.

(4) Genetic Improvement of Product Quality and Value

Selected Accomplishments: Progress has been made by ARS researchers in the identification and development of germplasm having enhanced health benefits and chemoprotective properties for consumers. Examples include broccoli inbreds and hybrids with higher concentrations of glucosinolate glucoraphanin (42), high beta-carotene tomato germplasm lines, and potato and carrot germplasm with health-promoting pigments.

Value-added traits were introgressed into ARS germplasm of nursery crops including: wheat (enhanced milling and bread-making attributes), soybean (enhanced yield potential), cotton (superior fiber and day-neutrality), sunflower (modified fatty acid profile), oat (higher soluble fiber content), and tomato (enhanced nutritive value) (24, 29, 30). HiFi, an oat cultivar released in 2001, has superior agronomic characteristics and a higher percentage of soluble fiber (6% beta-glucan) (30). Commercial oats (imported from Canada) average 4% beta-glucan. Other value-added traits include amylose-free spring wheat (43) and sunflower with a fatty acid profile very similar to olive oil (44).

The development of alternative crops include 'Spring Satin,' the first publicly released plumcot (plum-apricot hybrid), and the development of new and improved germplasm lines of guayule and lesquerella, which are being grown by commercial cooperators (46,47). 'Spring Satin' enables fruit growers to diversify their production. Lesquerella germplasm has been used by Terresolve Inc. for developing products from lesquerella oil for test marketing. Technology Crops International (TCI) has invested in a CRADA and is actively marketing lesquerella oil.

In efforts to reduce trans-fat and saturated fat in food products, companies such as McDonalds, Frito-Lay and Procter & Gamble use oil from NuSun, a new ARS-developed type of sunflower with high-oleic acid content. NuSun hybrids also were created with lower saturated fat content (48,49). In addition, ARS has led USB and QualiSoy sponsored efforts to develop soybeans with lower linolenic acid, lower saturated fat, and the first non-transgenic high-oleic soybean germplasm. These oil traits reduce the need for processing (hydrogenation) for food use, and provide vegetable oil processors and food manufacturers added flexibility for achieving better flavor, improved frying stability, and healthier food products.

New ARS white winter wheat cultivars adapted to the Great Plains may be used in the establishment of a new market class of winter wheat (50,51), with a concomitant increase in demand for U.S. hard wheat both in export and domestic markets. Spring wheat breeders now have access to the first waxy (amylose-free) germplasm adapted to Great Plains environments from an ARS research program (43). These germplasm lines will form the basis for the creation of yet another market class of wheat. The U.S. food industry now will have a novel raw material from which improved specialty flours and modified food starches may be created.

Value added phytochemicals that can positively impact consumer health and therefore increase demand for established crops have been identified by ARS researchers. Germplasm with increased concentrations of these beneficial phytochemicals have been released in broccoli, potato, and tomato. In the case of tomato, high beta-carotene tomato germplasm lines released by ARS have been utilized by cooperators at one large tomato processing company to develop proprietary breeding lines and by cooperators at another tomato processor to develop a nutritionally enhanced tomato sauce product for retail sale. These breeding lines are also being utilized by the Asian Vegetable Research and Development Center to develop locally adapted beta-carotene-rich cultivars to address endemic vitamin A deficiency in populations of Southeast Asia.

Guayule, a shrub native to the southwestern United States, represents a domestic source of natural rubber. Latex rubber from guayule is hypoallergenic, creating very high demand products for the medical care industry. Improved germplasm of guayule supplied by ARS to Yulex (a company commercializing guayule) has allowed the company to meet their goal of planting over 3,000 acres of guayule by the end of 2005. Latex produced from the production of guayule by Yulex will marketed by an international company. Signed contracts for latex will cause the expansion of production to about 75,000 acres within a few years. Guayule has the potential to be one of the most commercially successful new crops in the United States since the introduction of soybean.

Impact: The development of novel traits in existing crops and the development of alternative crops for producers can result in new market opportunities, new production opportunities, increase in returns for growers, and new products for consumers. As agricultural production systems become larger and more uniform, they also become more vulnerable to disasters such as drought or disease. Crop diversification not only provides increased returns for growers and new products for consumers, but it also provides stability to rural areas by reducing economic vulnerability. For example, the pending impact of guayule on the Southwest is especially important to replace the lost income from declining cotton production, particularly on Indian reservation land.

(5) New Genetic Methods for Crop Enhancement

Selected Accomplishments: ARS scientists have created technologies for crop genetic enhancement that range from biotechnology applications to on-farm organic agriculture research applications. Biotechnology methods include the development of a Plum Pox virus-resistant transgenic plum variety 'HoneySweet' and other stone fruits species (52). A gene for late-blight resistance was identified in wild diploid potato, cloned, sequenced, and transformed into cultivated potato where resistance was demonstrated to be strong (53). ARS researchers have also developed a unique system for inducing and normalizing somatic embryogenesis in citrus cell lines (54).

ARS researchers applied DNA markers to enhance germplasm via marker-assisted selection breeding strategies in numerous species. Quantitative inherited traits such as disease resistance, flowering, plant height, salt-tolerance, seed dormancy, rhizome proliferation, forage mineral content, grain cooking quality may now be targeted for enhancement simultaneously in segregating populations of rice, potato, lettuce, chickpea, and grasses (55). DNA marker analysis of ornamentals has been used to estimate diversity within genera, verify interspecific and intraspecific hybrids, and DNA-fingerprint cultivars and varieties for unambiguous identification (56). Likewise, numerous new common bean germplasm lines with improved disease resistance have resulted from use of the different marker-assisted selection strategies developed by ARS researchers.

Breeding programs have also been aided by the development of phenotypic assays for traits that have previously been difficult, time-consuming, or expensive to evaluate. For example, rapid and high throughput methods to measure nutritionally limiting nutrients such as lysine, tryptophan, methionine and phytic acid in maize grain were developed (57). Maize breeders in

the public and private sectors adopted these methods to develop varieties with improved nutritional quality. Similarly, ARS scientists identified the key end-product quality traits of wheat, documented the underlying genetic basis for their variation, and developed efficient methods to screen large numbers of germplasm lines (58). A simple germination stress test for sugarbeet using seeds germinated in water was developed and was found to predict field emergence, an important and difficult to select trait (59). The test is able to discriminate variety seedling vigor, as well as differences in vigor among different seedlots of the same variety. Finally, an ARS research group developed a method to rapidly convert elite sunflower lines from low- to high-oleic fatty acid utilizing gas chromatography to assay fatty acid profiles and embryo culture to speed generation times. Four to five generations of seed have been advanced in one year utilizing this method (60).

ARS research on reproductive biology of crops has resulted in several major advances in the past five years. For example, data on self-compatibility and time of stigma receptivity have improved the effectiveness of controlled pollinations in *Hydrangea*, *Styrax* and *Cornus* (61,62). This research has improved the efficiency of plant improvement in these woody ornamental species. Additionally, in soybean, the difficulty in making sexual crosses has limited soybean to an inbreeding crop. ARS scientists have developed a hybrid seed production procedure that entails phenotypic recurrent selection and to increase insect pollinator activity (63). This research has yielded a five-fold increase in out-crossed seed set.

The development of improved phenotypic assays for difficult traits will open up new breeding and research opportunities. For example, simple assays, such as the L-DOPA PPO assay for improved wheat flour color, facilitate the efficient screening of large breeding populations and are being used throughout the United States and around the world. Similarly, the tools and strategies developed to rapidly assay maize grain nutritional quality have been adopted by several researchers who previously did not work on nutritional quality because of the lack of easy assays. Because the nutritional quality of maize in turn affects the cost of meat production, this research could lead to reduced costs of food production.

The development of a procedure to produce large numbers of hybrid soybean seed would dramatically change the dynamic of soybean breeding. A large increase in hybrid seed production would permit more parental combinations to be evaluated in breeding programs. Indeed, industry is already adapting this procedure to produce large numbers of hybrid seed to increase locations and replications in a larger number of hybrid combinations.

Finally, ARS research on the improvement of breeding methods included on-farm maize variety evaluations, which were initiated with participating farmers (Practical Farmers of Iowa). The farmers take an active role in helping ARS researchers manage and evaluate the experiments, and some participate in participatory breeding by adapting populations to their farms. A breeding nursery was established in cooperation with an organic producer to conduct maize selection experiments for adaptation to organic production. Similar projects have been initiated for vegetable crops.

Impact: ARS has pioneered new methods and technologies for genetic improvement of agricultural crops that have facilitated the development of value-added germplasm that might

otherwise be too difficult or expensive to create. In addition, the application of genomic technology has ushered in a new era in marker-assisted selection programs that enables the simultaneous efficient selection of progeny exhibiting specific genes for multiple traits of interest for enhanced productivity and crop quality.

(6) Statistical Approaches to Optimize Breeding Progress

Selected Accomplishments: ARS researchers have conducted both basic and applied research on statistical genetics as applied to crop improvement in the past five years. Examples of basic research on statistical genetics are new association mapping methods that reduce the rate of false positive associations between candidate genes and phenotypes (64), and new methods for estimating heritability and genetic correlations that are robust to missing data (65).

Applied statistical genetic research, heritabilities and genetic correlations of economic traits were identified in the extensive USDA-ARS hop germplasm collection (66). Classical mating studies and molecular genetic approaches were also used to identify potential heterotic groups and subsets in this collection. Similarly, the heritability and stability of internal necrosis and specific gravity in potato were analyzed by ARS scientists. Another example is the analysis of the National Cotton Variety Test database. ARS manages the National Cotton Variety Test, involving 15 states and 38 locations, thus ensuring consistent and unbiased testing and reporting (67), and also providing a unique opportunity for statistical analysis of key agronomic traits in cotton. Investigations over years and locations showed that about 80% of variability in cotton for yield and fiber color is due to environmental conditions. Conversely, the environment accounts for only 50% of the variability for fiber strength and length.

Impact: New statistical genetics methods developed by ARS researchers have been used to conduct improved genetic analyses and have been adopted by outside researchers. The new association mapping techniques were developed and applied to identify specific genes underlying flowering and grain quality traits in maize (68). The new heritability and correlation estimation procedures have been used in a number of crops, increasing the reliability of statistical genetic studies. Software for these methods was made freely available on the web (69,70).

The results of applied statistical genetic studies guide breeding programs by determining the optimal breeding methods for traits of different levels of heritability and stability. Having accurate estimates of trait heritabilities helps breeders to optimally allocate their resources to maximize genetic gain (for example, by determining a minimum number of testing environments needed to make selections).

RESEARCH COMPONENT III: GENOME DATABASES AND BIOINFORMATICS

Five years ago, this Research Component called for constructing and maintaining crop genome databases, and for developing bioinformatic tools essential for effectively managing new genetic information being generated at an increasingly rapid rate. Bioinformatic software for effectively storing, processing, and organizing DNA sequence and microarray information were particularly important to produce and continually update. Knowledge gained from genomic analyses of

model species would be applied to accelerate crop genome analyses. Infrastructure would be developed to provide collaborative links, through computer networks, among ARS laboratories, other public sector programs, and private sector research efforts.

Through joint planning and collaboration with research partners, USDA/ARS essentially sought to catalyze the evolution of "crop genome research communities" which would be instrumental for accelerating progress in crop genetics and genetic improvement for the 21st Century. During the past five years, USDA/ARS crop genome database development was coordinated with plant genome sequencing and characterization projects supported by other Federal agencies. Particularly through the Interagency Working Group on Plant Genomes (http://www.ostp.gov/NSTC/html/npgi05/interagency.htm), USDA/ARS has cooperated on developing crop genome databases and bioinformatics tools with the NSF Plant Genome Program, and the plant genome/genetics programs of USDA/CSREES, DOE, and NIH.

Problem Area IIIa: Long-term Stewardship of Genome Databases

Outcomes, Goals, and Commitments: USDA/ARS committed to maintaining and refining crop genome databases for the long-term. Attaining that goal required that databases were routinely updated via data acquisition and data entry. Stable, state-of-the-art, high quality databases and database management capabilities had to furnish ready access to a broad diversity of data. A common set of query tools was required to evaluate data relationships efficiently. Data models required revision and refinement so as to have more features in common. Genome databases had to take into account novel relationships among data sets, and facilitate cross-species comparative studies. Database structure and function required continual modification so as to incorporate data generated by new genetic marker technologies for genotyping, functional genomic data, etc.

Not only did USDA/ARS commit to database stewardship per se, but also to developing database access mechanisms that were faster, or that provided increased functionality and utility to the user. New or improved standards for genome database curation had to be developed and implemented. Technical impediments to long-term stewardship of genome databases had to be removed.

Selected Accomplishments: Key genome databases for several major crops (soybean, maize, and wheat/barley/oats) were maintained effectively during the last five years. New database models for these crops that encompass new technologies and data types were developed and implemented. For example, five years ago, extensive genomic sequence data did not exist for most crops. Today, the large quantities of sequence information furnish a common language for inter-database communications. Five years ago, functional genomics was in its infancy. Today, gene expression information constitutes an integral connection between genetic data and phenotypic data. USDA/ARS crop genome databases evolved to safeguard and deliver, 24/7, those new data to researchers worldwide. In addition to providing access to genomic sequence data, and beginning to develop data models for expression data, database models were updated for metabolic pathways, cell lines and transgenes, pathogens, germplasm, and new genetic marker types.

Genetic maps constitute invaluable tools for both genomics and breeding, and should also serve as key practical and conceptual links between the latter fields of research. Genetic maps constructed from multiple populations and nearly 4,000 markers were interpolated into a single composite map for soybean. To make this map information pertinent to breeders, QTL from dozens of mapping studies were included. This composite map was added to Soybase (1) and to the cross-species legume database (LIS, the Legume Information System, 2). Many of the soybean markers proved applicable to genomic research with other legumes, especially to the model legume species, *Medicago truncatula* (3).

NP 301 scientists significantly enhanced maize databases by curating genetic mapping data that include more than 7,500 mapped loci, and adding another 20,000 "overgos" to the maize genetic framework (4). The Gramene (5) and GrainGenes (6) databases expanded to include community database genetic maps, and protein, gene expression and phenotypic data, to name but a few data types. These databases facilitated research with the grasses in general, but particularly with wheat, barley and oats. Through a cooperative endeavor with the National Center for Genomic Research, new models and software were created to facilitate cross-species comparisons through the LIS.

> **Impact:** Because of the preceding achievements, researchers now have rapid access to critical, biologically relevant genetic data for many crops. Dataset and database design now enable cross-species comparisons, so that advances in one crop may accelerate research progress in others. The long-term stewardship of crop databases by NP 301 personnel resulted in SoyBase, the LIS, and GrainGenes becoming vital components of crop genomics infrastructure, ensuring that researchers could exploit up-to-date data representing the cutting edge of science. SoyBase and LIS became critical components of all the Phaseoloid genomic efforts. GrainGenes provided informatics support for large-scale genomics efforts in wheat, barley and oats. These databases helped to minimize redundancy in research and to provide a ready source of information for students, researchers and the general public.

Problem Area IIIb: Development of Interconnected and Interoperable Genome Databases

Outcomes, Goals, and Commitments: USDA/ARS committed to initiating, developing, implementing, or adopting new interoperable and interconnected genome databases with consistent data models and user interfaces. Powerful, interoperable queries were required for comparative genomic mapping, structural genomic, functional genomic, and proteomic research. Furthermore, automated procedures were needed to cross-relate information among widely diverse sources of data, and improve artificial intelligence, interoperability, data mining, and cross-querying capabilities. Genome databases required harmonized, controlled vocabularies for plant metabolism, etc. and, in conjunction with strong user input, precise, deep-searching, and intelligent data mining/recovery systems had to be specially tailored to analyzing crops. Genome database (especially data structuring) and bioinformatic tool development in individual databases had to be coordinated with one another, and with data management efforts for genetic resource curation (see Research Component I), structural and functional genomics, and genetic improvement (see Research Component II).

Selected Accomplishments: The advent of the genomic era created an ever-increasing volume of genetic data. Crop databases rose to the challenge of managing those data. Bioinformatic resources were developed and implemented to increase the interoperability and interconnectivity of databases. These included new database structures, visualization and analysis tools, controlled vocabularies, and new community databases per se.

During the last five years, USDA/ARS and its research partners made significant progress in adopting relational database structures to crop genome databases, developing user-friendly web interfaces, and in constructing new bioinformatics tools for researchers. In 2003, the earliest maize genome database, MaizeDB, was transformed into a "second-generation" database (9), Maize GDB (Maize Genetics and Genomics Database). Data in ZmDB, another early maize genome database, were also incorporated into MaizeGDB. In 2004, the earlier GrainGenes database completed the migration of all GrainGenes data (in MySQL) into "GrainGenes 2.0", a relational database (12) This new relational database enabled cereal researchers and breeders to apply tools and interfaces from other genome database to analyze data maintained in GrainGenes 2.0 (13).

Gramene (1), a comparative grass genomics database, was developed during the last five years, and it now enables the completed sequence of rice genome to serve as a reference tool for cereal genetics, genomics and breeding as a whole. Similarly, the LIS (2), a community database for the legumes, enables *Medicago, Lotus and Arabidopsis* genomic data to serve as a research tool for those economically-important plants.

Gramene and SoyBase (3) personnel actively collaborated with the broader plant research community to establish controlled vocabularies and foster cross comparative analyses and database interoperability (4). These databases provided internal and external links to other public databases that facilitated cross-referencing and browsing within and between different datasets and databases. SoyBase provided direct connections to the LIS, as well as to *Medicago* databases. Gramene contributed directly to the Generic Model Organism Database project ("GMOD", an open source software repository for application tools) (5). All the crop databases developed unique tools, but also readily exploited open-source bioinformatics software so as to minimize redundant efforts.

> **Impact:** Five years ago, almost all crop databases were `stand alone' entities, in non-relational formats, with no hopes of inter-communicating. As a result of research within this Problem Area, the research community now has several interoperable databases for key agronomic crops. MaizeGDB (6, 9), a sequence-based database, now serves as a central clearinghouse for maize genetic information. Gramene provides a similar resource for other grasses (7), GrainGenes for wheat, barley, and oats (8), SoyBase/LIS for the legumes (10), and CocoaGenDB (via an USDA/ARS-international consortium) for cacao (11).
>
> The interconnectivity of crop databases now enables research breakthroughs in one species to be immediately applied to genomic analyses of other species. This accelerated the flow of knowledge throughout research communities. As an example, the Gramene contribution of CMap to GMOD (5) enabled ready comparative alignment of genetic

maps for a variety of species. Better characterization of plant genome structure, organization, and gene content accelerated the identification of genotypes associated with agronomically favorable traits in both moncots and dicots. More powerful comparative genomic analyses and gene prediction programs aided crop improvement programs.

Problem Area IIIc: Analyses of Genomic Data

Outcomes, Goals, and Commitments: Under this Problem Area, USDA/ARS sought to develop new bioinformatic approaches, standards, and tools for elucidating the genetic bases for complex traits, such as pathogen-host interactions and response to environmental stresses. New approaches had to be formulated for analyzing genome diversity, and specific types of sequence and gene expression data. Analyzing these data types effectively called for new types of data representation, database design, visualization, and a means for assessing the association between genetic resource evaluation and geographic data.

Selected Accomplishments: Several tools that aid in the analysis and presentation of new genomic data were developed during the last five years. For example, research at Ames, IA created a suite of tools that can analyze EST data from public databases and identify members of gene families, as well as single nucleotide polymorphisms (SNPs) among individual genes (1). Analyses of extensive EST data from numerous crops revealed genomic polyploidy events that occurred during the crop's evolution (2). Research at Columbia, MO produced the IBM neighbors map resources that serve as the current "scaffold" for the maize genome sequencing effort (3). This scaffold contains close to 30,000 map sites, plus 20,000 genes ("overgos") and SSRs.

Impacts: The massive volumes of genomic data that flowed into crop databases during the last five years could be deployed for research only if they could be readily accessed, queried, sorted and analyzed. The software tools constructed during the last five years helped attain the preceding goals, thereby enabling those data to be exploited by the crop research community. For example, developing the capability for high through-put SNP identification alleviated a bottleneck in genetic marker development, thereby accelerating the identification of molecular markers that breeders can apply to crop improvement. EST data analyses discerned major evolutionary relationships among crops and generated valuable information about genome relatedness, without the necessity of sequencing entire genomes. Ready access to the IBM neighbors map and placement of thousands of markers on that map will accelerate progress with the maize genome sequencing project, as well as enabling more efficient identification of candidate genes for major agronomic traits.

APPENDIX 1 - SELECTED SUPPORTING INFORMATION AND DOCUMENTATION FOR ACCOMPLISHMENTS AND IMPACT OF NP301 RESEARCH

Planning and Coordination for NP301

1. National Workshops
    - NP 301 Customer and Stakeholder Workshop, 2000
    - Plant Germplasm Operations Committee Annual Meetings, 2000-2005
    - Crop Germplasm Committee Chairs Biennial Meetings in 2000, 2002, and 2004
    - National Fusarium Head Blight Annual Forums, 2000-2005
    - USB-ARS QualiSoy Initiative and Seed Composition Annual Workshops, 2000-2005
    - ARS Rice Research Workshop, 2001
    - Comparative Insect Genomics Workshop, 2001
    - ARS Sclerotinia Strategic Planning & Research Forums, 2002-2005
    - ARS-USB Soybean Genomics Workshops 2002-2005
    - ARS and Private Sector Sorghum Research Workshop, 2003
    - ARS Floriculture & Nursery Research Initiative Researchers Meeting, 2003
    - National Citrus Genomics Workshops, 2003-2005
    - Joint NSF-USDA/ARS site visits and database reviews for MaizeDB (2003), MaizeGDB (2004)
    - Technology Roadmap Temperate Fruit Genomics, Genetics, and Breeding Workshop, 2004
    - ARS-APC Peanut Genome Workshops, 2004-2005
    - International Sunflower Research Conference, 2004
    - National Soybean Rust Research Workshop, 2004
    - Legume Crop Genome Initiative Workshop, 2004
    - ARS-USB Plant-based feeds in Aquaculture Workshop, 2005
    - ARS Wine/Grape Industry Workshop, July 2005

Component I: Genetic Resource Management

Problem Area Ia: Safeguarding Threatened Genetic Resources and Associated Information

1. Williams, K.A. 2005. An overview of the U.S. National Plant Germplasm System's Exploration Program. Hortscience 40:297-301.

2. In the last five years, plant explorations have occurred in Armenia, Azerbaijan, Chile, Georgia, Greenland, Guatemala, Honduras, Italy, Japan, Kazakhstan, Mexico, Nicaragua, Palestine, Panama, Paraguay, People's Republic of China, Portugal, Russian Federation, Samoa, South Georgia, Tahiti, Tajikistan, Turkey, Turkmenistan, Uzbekistan, and the United States.

3. In the last five years, germplasm exchanges were conducted with Israel, Palestine, Jordan, Tunisia, India, Bangladesh, Sri Lanka, Pakistan, Uzbekistan, Turkmenistan, Tajikistan, Kyrgyzstan, Kazakhstan, Armenia, Azerbaijan, Georgia, Cameroon, Ecuador, Guatemala, Mexico, Peru, Bolivia, and Paraguay.

4. Crops collected in plant explorations in the last five years included wheat, barley, garlic, onion, carrot, pea, tomato, chickpea, lentil, arugula, chile pepper, collard, hops, spinach, rangeland legumes, peanut, spices, sweet potato, sunflower, cotton, rice, tomatillo, breadfruit, fruits, nuts, grasses, and ornamentals.

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Problem Area Ib: Conserving Genetic Resources and Associated Information Efficiently and Effectively

1. http://www.ars-grin.gov/npgs/stats/summary.stats

2. Since 1989, over 3,000 genetically diverse rhizobial strains have been added to the collection.

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7. Original USDA accession, USDA 110, a rhizobial strain for soybean. 2002.

Problem Area Ic: Documenting and Characterizing Genetic Resources

1. Germplasm Resources Information Network, http://www.ars-grin.gov/npgs/

2. SINGER = Systemwide Information Network for Genetic Resources, http://singer.cgiar.org

3. From 2000-present, more than 36,000 new Accession Records, more than 280,000 new Inventory Records, more than 310,000 new Viability Records, more than 1,700,000 new

Observation Records (essentially, characterization and evaluation data), as well as more than 26,000 Orders and almost 800,000 Order Items were incorporated into GRIN.

4. Jarret, R.L, Perkins, B., Fan, T., Prince, A., Guthrie K., Skoczenski, B. 2003. Using EIA to screen *Capsicum* germplasm for capsaicinoid content. Journal of Food Composition and Analysis. v. 16. p. 189-194.

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6. Spooner, D.M., van den Berg, R.G., Rodríguez, A., Bamberg, J., Hijmans, R.J., Lara-Cabrera, S.I. 2004. Wild potatoes (*Solanum* section *Petota*) of North and Central America. Syst. Bot. Monogr. v. 68. p. 1-209 + 9 plates.

7. Spooner, D.M., Hijmans, R.J. 2001. Potato systematics and germplasm collecting, 1989-2000. Amer. J. Potato Res. v. 78. p. 237-268; 395.

Problem Area Id: Expanding Germplasm Evaluations and Characterizations

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2. Grauke, L. J., Iqbal, M. J., Reddy, A. S., and Thompson, T. E. 2003. Development of microsatellite DNA markers in pecan. J. Amer. Soc. Hort. Sci. 128(3):374-380.

3. Saunders, J., Mischke S., Leamy, E.A. and Hemeida A.A. 2005. Selection of International molecular standard for DNA fingerprinting. Theoretical and Applied Genetics. v. 110. p. 41-47.

4. Schnell, R. J., J. S Brown, C. T. Olano, E. J Power, C. A. Krol, D. N. Kuhn, and J. C. Motamayor. 2003. Evaluation of Avocado Germplasm Using Microsatellite Markers. J. Amer. Soc. Hort. Sci. 128(6):881-889.

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Problem Area Ie: Technology Transfer of Genetic Resources and Associated Information

1. Bowman, J.G.P, Blake, T.K., Surber, L.M.M., Habernicht, D.K., Bockelman, H.E. 2001. Feed-quality variation in the barley core collection of the USDA National Small Grains Collection. Crop Sci. 41:863-870.

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Component II: Genome Characterization and Genetic Improvement

Problem Area IIa: Genome Characterization

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Problem Area IIb: Genome Improvement

1. ARS research has resulted in the release of cultivars of the following species: Grains: wheat, barley, rice, and oat; vegetables: potato, sweetpotato; legumes: soybean, chickpea, dry bean, dry pea, lentil, and Southern pea (cowpea or blackeye bean); fruits/nuts: apple, blackberry, grape, peach, pear, pecan, plum, plumcot, raspberry, and strawberry; forage/range grass: buffelgrass, eastern gamagrass, dallisgrass, and sand bluestem; ornamental: American elm, *Capsicum* (ornamental), *Clethra*, crapemyrtle, flowering cherry, *Hemerocallis, Hydrangea*, lilac, *Iochroma, Polygala myrtifolia*, redbud, and Star-of-Bethlehem (*Ornithogalum*); sugar: sugarcane.

2. ARS research has resulted in the release of enhances germplasm of the following species: maize, wheat, sunflower, sugarbeet, grain sorghum, broccoli, carrot, cucumber, common and dry bean, lettuce, potato, tomato, switchgrass, wheatgrass, wildrye, ricegrass, needlegrass, bromegrass, bottlebrush squirreltail, and bluegrass.

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Component III: Genome Databases and Bioinformatics

Problem Area IIIa: Long-term Stewardship of Genome Databases

1. http://soybase.org

2. The Legume Information System was cooperatively developed between ARS and the National Center for Genomic Research, Santa Fe, NM; http://www.comparative-legumes.org

3. http://www.medicago.org

4. http://www.maizeGDB.org

5. http://www.Gramene.org

6. GrainGenes 2.0 at http://wheat.pw.usda.gov/GG2/index.shtml

Problem Area IIIb: Development of Interconnected and Interoperable Genome Databases Supporting Information and Documentation

1. http://www.Gramene.org

2. http://www.comparative-legumes.org

3. http://soybase.org

4. http://www.plantontology.org

5. http://www.gmod.org

6. http://www.maizeGDB.org

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Problem Area IIIc - Analysis of Genomic Data

1. Nelson, R.T., Grant, D., Shoemaker, R.C. 2004. ESTminer: A Tool for Identifying Locus and Allele Defining Polymorphisms Using EST Libraries. Bioinformatics. 21:691-693.

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3. Polacco, M.L., Sanchez-Villeda, H., Coe, E.H.,Jr. 2003. A Consensus Map: Inter-mated B73 x Mo17 (IBM) Neighbors. Maize Genetics Cooperation Newsletter v. 77. p. 137-179.

Appendix 2 – Research Projects

Component I

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
Alaska			
	Arctic Plant Germplasm Research and Introduction	A. Pantoja (P) D. Ianson N. Robertson	1/28/03
California			
	Management of Fruit and Nut Crop Genetic Resources	E. Stover (P) M. Aradhya	5/16/03
	Arid Land Plant Genetic Resources Conservation, Evaluation and Information Management	M. Jenderek (P) R. Hannan	6/11/03
	Citrus and Date Genetic Resources and Information Management	R. Lee (P) R. Krueger	4/26/05
Colorado			
	Preservation and Quality Assessment of Plant Genetic Resources	D. Ellis (P) H. Shands	8/5/03
	Research to Develop Strategies and Technologies for Preserving Plant Genetic Diversity in Ex Situ Genebanks	C. Walters (P) L. Towill G. Volk C. Richards	10/17/03
Florida			
	Conservation, Characterization, and Genetic Improvement of Subtropical and Tropical Ornamental Germplasm	A. Meerow (P)	6/18/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Conservation and Utilization of Subtropical/Tropical Fruit Crops, Sugarcane and Tripsacum Genetic Resources	R. Schnell (P) T. Ayala J. Brown	12/17/03
Hawaii			
	Assuring Long Term Preservation of Tropical Crops Via Genetic Resource Management	F. Zee (P) D. Gonsalves L. Keith B. Matsumoto	9/6/03
Idaho			
	Small Grains Genetic Resource Management	H. Bockelman (P) J. Bonman	6/18/03
Illinois			
	Management and Genetic Characterization of Agricultural and Biotechnological Microbial Resources	D. Labeda (P) S. Peterson C. Kurtzman K. O'Donnell T. Ward A. Rooney	6/16/04
	Soybean Genetic Resources Management and Utilization	S. Clough (P) R. Nelson G. Hartman	8/1/03
	Conservation and Utilization of Maize Genetic Stocks	M. Sachs (P)	6/18/03
Iowa			
	Plant Genetic Resource and Information Management	C. Gardner (P) C. Block M. Widrlechner	2/8/04
Maryland			

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Systematics, Nomenclature, and Genetic Diversity Assessment Research to Facilitate the Use of Vascular Plants as Genetic Resources	J. Kirkbride (P) J. Wiersema	6/18/03
	Federal Quarantine for International and Domestic Movement of Saccharum Genetic Resources	S. Hurtt (P) R. Li F. Hammerschlag	5/1/03
	Federal Quarantine for the Importation of Plant Genetic Resources	G. Kinard (P) S. Hurtt R. Li F. Hammerschlag R. Mock	4/17/03
	Plant Explorations to Acquire Crop Genetic Resources	K. Williams (P) E. Garvey	5/2/03
	Exchange of Crop Genetic Resources and Associated Documentation for the U.S. National Plant Germplasm System	E. Garvey (P) K. Williams R. Webster	4/11/03
	Operating and Developing the Germplasm Resources Information Network for the U.S. National Genetic Resources Program	J. Mowder	5/7/03
	Development of On-Line Systematic Resources About Fungi and the U.S. National Fungus Collections	D. Farr (P) A. Rossman	6/18/03
	Preservation of Honey Bee Germplasm	A. Collins (P) J. Evans	9/10/03
	Exotic Pathogens of Citrus	J. Hartung (P) F. Hammerschlag	3/27/03
New York			
	Conservation and Utilization of the Genetic Resources of Apples, Grapes, and Tart Cherries	P. Forsline (P) C. Simon A. Baldo	8/7/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Conservation and Utilization of Germplasm of Selected Vegetable Crops	L. Robertson (P) J. Labate A. Baldo	9/13/03
	Bioanalytical Methods and Tools for Small Grains	P. Bradbury	3/21/05
	Microbial and Genetic Resources for Biological Control and High-Value Uses	D. Gibson (P) R. Humber	6/1/04
Oregon			
	Management of Temperate Adapted Fruit, Nut, and Specialty Crop Genetic Resources and Associated Information	K. Hummer (P) N. Basil J. Postman B. Reed	6/6/03
Puerto Rico			
	Conservation, Management and Genetic Enhancement of Sorghum Genetic Resources and Associated Information	J. Erpelding (P)	1/7/04
	Genetic Resource Management of Tropica/Subtropical Genetic Resources and Associated Information	B. Irish (P) R. Goenaga	2/1/05
	Seed Increase and Phytosanitary Assessment of Quarantined and Tropically-Adapted Genetic Resources	R. Goenaga (P)	2/26/04
Texas			
	Conservation and Characterization of Genetic Diversity in Carya	L. Grauke (P) T. Thompson	6/18/03
Washington			
Pullman			
	Plant Genetic Resources Conservation, Research and Information Management	R. Hannan (P) B. Hellier F. Dugan V. Bradley T. Kisha S. Clement J. Coyne M. Welsh R. Johnson	8/5/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Temperate Forage Legume Genetic Resources Conservation and Information Management	S. Greene (P) R. Hannan	1/29/03
Washington, DC			
	Taxonomy and Genetic Diversity Assessment of Landscape Trees and Shrubs	A. Whittemore (P)	8/5/03
	Education and Visitor Services at the U.S. National Arboretum	N. Luria (P)	12/1/02
	Genetic Resources, Evaluation and Information Management of Woody Landscape Plant Germplasm	M. Roh (P)	6/11/04
	Establish Public Display Gardens for Woody and Herbaceous Landscape Plants	S. Aker (P) T. Elias	6/27/01
Wisconsin			
Madison			
	Systematics, Genetic Diversity Assessment, and Acquisition of Potatoes and Related Wild Relatives	D. Spooner (P) P. Simon	7/16/03
	Conservation and Utilization of Potato Genetic Resources	J. Bamberg (P) P. Simon	11/19/03

Component II

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
Arizona			
Phoenix			
	Evaluation, Improvement, and Development of New/Alternative Industrial Crops	T. Coffelt (P) D. Dierig	7/24/03
	Physiological and Genetic Basis of Cotton Acclimation to Abiotic and Biotic Stress	S. Crafts-Brandner (P) B. Deridder R. Percy M. Salvucci	4/25/01
Arkansas			

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
Stuttgart			
	Use of Diverse Germplasm for Genetic Improvement of Rice	J. Rutger (P) W. Yan R. Bryant G. Eizenga	8/1/03
	Genomic Characterization of Rice Germplasm	S. Brooks (P) G. Eizenga N. Rutger Y. Jia	6/10/04
Alaska			
Fairbanks			
	Development of Virus-Free Potato Germplasm	A. Pantoja (P)	9/9/02
California			
Parlier			
	Improvement of Prunus and Vitis Scion and Rootstocks for Fruit Quality and Pest Resistance	C. Ledbetter (P) D. Ramming	8/1/03
Salinas			
	Evaluation, Enhancement, Genetics, and Breeding of Lettuce, Spinach, Melon, and Closely Related Species	J. McCreight (P) B. Mou R. Hayes	7/27/03
	Genetics, Breeding and Enhancement of Sugarbeet Germplasm for Resistance and Productivity	R. Lewellen (P)	7/19/03
Davis			
	Rice Genetics and Germplasm Enhancement for Temperate Environments	T. Tai (P)	6/18/03
Albany			
	Improved Molecular Genetic Tools for Potato Improvement	W. Belknap (P) K. McCue	8/17/03
	Molecular Basis of Key Cereal Grain Traits	O. Anderson (P) D. Chingcuanco Q. Gu	7/24/03
	Crop Improvement through Directed Genetic Recombination	D. Ow (P)	3/27/01
	Functional Genomics of Plant Architecture	J. Fletcher (P)	4/6/01

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Molecular Mechanisms of Light Perception, Signaling, and Control of Gene Expression by the Phytochromes	S. Hake (P)	10/1/00
	Molecular and Genetic Analyses of Pollen Development and Pollen-Pistil Signaling in Crop Plants	S. McCormick (P)	3/17/01
Shafter			
	Integrated Management of Pests Affecting Cotton: Plant Genetics, Biocontrol, and Novel Methods of Pest Estimation	M. McGuire (P) J. Bancroft M. Ulloa	5/25/05
Colorado			
Fort Collins			
	Nonchemical Pest Control and Enhanced Sugarbeet Germplasm via Traditional and Molecular Technologies	L. Panella (P) L. Hanson R. Larson	7/27/03
Florida			
Fort Pierce			
	Genetic Improvement of Citrus	K. Bowman (P) M. Bausher R. Niedz T. McCollum	5/2/03
Canal Point			
	Enhancement of Sugarcane Germplasm for Development of Cultivars and Sustainable Production	S. Edme B. Glaz	7/1/03
Miami			
	Development of DNA Markers Associated with Disease Resistance in Cacao	R. Schnell (P) J. Brown T. Ayala	4/18/03
Gainesville			
	Biologically-Based Technologies for Management Crop Insect Pests in Local and Areawide Programs	J. Sivinski (P) R. Meagher R. Nagoshi A. Handler P. Shirk R. Mankin	7/20/05
Georgia			
Griffin			

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Conservation, Characterization, and Evaluation of Crop Genetic Resources and Associated Information	G. Pederson (P) A. Gillaspie R. Jarrett B. Morris R. Pittman M. Newman L. Wang	9/27/03
Tifton			
	Development of Improved Peanut Germplasm with Resistance to Disease and Nematode Pests	C. Holbrook (P)	5/22/03
	Genetic Improvement of Corn and Sorghum for Resistance to Insects and Aflatoxin	M. Krakowsky (P) X. Ni C. Holbrook, Jr.	8/1/2005
	Integration of Alternative Pest Management Strategies for Management of Insects and Aflatoxin Contamination	J. Carpenter (P) B. Guo W. Lewis	1/28/03
Byron			
	Deciduous Fruit Improvement and Protection from Pests	W. Okie (P) T. Cottrell T. Beckman	4/24/03
Hawaii			
Hilo			
	Genomic and Biotechnological Approaches for Evaluation and Improving Tropical Crops	P. Moore (P) H. Albert D. Gonsalves	9/27/03
Idaho			
Aberdeen			
	Seed Chemistry Genetics	V. Raboy (P) J. Bonman	5/9/08
	Barley and Oat Germplasm Evaluation and Enhancement	J. Bonman (P) A. Hang D. Hoffman D. Obert P. Bregitzer	7/6/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Development of Potato Varieties and Germplasm with Improved Yield, Quality, Disease and Pest Resistance	R. Novy (P) J. Bonman	7/1/03
Kimberly			
	Enhancement of Sugarbeet Germplasm for Improved Resistance and Productivity	C. Strausbaugh (P) A. Gillen	5/5/03
Illinois			
Peoria			
	Coordinated Analysis of Soybean Breeding Germplasm	T. Isbell (P)	4/1/01
Indiana			
West Lafayette			
	Identification and Characterization of Genes Important During Seed Development in Soybean	N. Nielsen (P)	3/20/01
	Enhancing Resistance to Root Rot Pathogens of Soybean	T. Abney (P) L. Dunkle	3/18/03
	Molecular and Genetic Mechanisms of Hessian Fly Resistance in Soft Winter Wheat	R. Shukle (P) C. Williams L. Dunkle B. Schemerhorn	6/18/03
Iowa			
Ames			
	Enhancing Agronomic and Value-Added Traits of Corn Germplasm	L. Pollak (P) L. Lewis	12/12/03
	Soybean and Pollinator Attraction	R. Palmer (P) L. Lewis	10/05/03
	Genetic Improvement of Maize as a Source of Protein Nutrition	M. Scott (P) L. Lewis J. Edwards	5/28/05

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Functional and Structural Genetic Analysis of Soybean	R. Shoemaker (P) L. Lewis	4/9/03
	Germplasm Enhancement of Maize Project (GEM)	M. Blanco (P) C. Garnder	7/24/04
	Breeding High-Quality Corn for Sustainable, Low-Input Farming Systems	L. Pollak (P) L. Lewis	4/9/04
Kansas			
Manhattan			
	Genomics and Proteomics of Stored-Product Insects for Development of New Biopesticides	R. Beeman (P) B. Oppert	4/6/05
	Genetic Enhancement for Resistance to Biotic and Abiotic Stresses in Hard Winter Wheat	R. Bowden (P) M. Chen J. Fellers G. Bai Z. Ristic	7/19/03
Louisiana			
Baton Rouge			
	Development and Use of Mite-Resistance Traits in Honey Bee Breeding	J. Harbo (P) J. Harris A. Sylvester R. Danka T. Rinderer	10/1/03
	Breeding, Genetics, Stock Improvement and Management of Russian Honey Bees for Mite Control and Pollination	R. Rinderer (P) L. De Guzman J. Villa A. Slyvester R. Danka	10/1/03
New Orleans			
	Genetic Improvement of Sugarcane by Conventional and Molecular Approaches	T. Tew (P) J. Veremis Y. Pan W. White E. Richard	8/1/04

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
Maryland			
Beltsville			
	Molecular Characterization and Diversity Assessment of Cocoa Germplasm in the Americas	B. Bailey (P)	6/25/04
	Genetic Enhancement of Potatoes for Nutritional and Processing Quality and for Resistance to Diseases	K. Haynes (P)	5/27/04
	Genetic Enhancement of Quality Constituents and Disease Resistance in Solanaceous Vegetables	J. Stommel (P)	7/24/03
	A Single Nucleotide Polymorphism-Based Map of Soybean and Applications to Gene Discovery in Germplasm	P. Cregan (P)	5/14/03
	Molecular Biology, Taxonomy, and Enhancement of the Bradyrhizobium Soybean Symbiosis	P. van Berkum (P) B. Cooper	1/1/01
	Enhancement of Small Fruit Germplasm through Genomic Characterization and Genetic Improvement with Emphasis on Disease Resistance	M. Ehlenfeldt (P) J. Polashock S. Wang K. Lewers F. Hammerschlag W. Turechek	6/5/04
	Reducing Genetic Vulnerability in Common Bean by Incorporating New Genes for Disease Resistance	M. Pastor Corrales (P)	7/19/03
	Enhancement of Blueberry And Strawberry Through Analysis and Modification of the Plant Genome	L. Rowland (P) R. Hammerschlag K. Lewers M. Ehlenfeldt J. Polashock	6/13/02
	Development and Utilization of Simple Sequence Repeat (SSR) Molecular Markers for the Improvement of Alfalfa and Related Species	R. Bauchan (P)	2/26/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Managing Diseases and Pests of Honey Bees to Improve Queen and Colony Health	M. Feldlaufer (P) J. Evans J. Pettis Y. Chen J. Kochansky	9/10/03
Michigan			
East Lansing			
	Breeding Selection and Molecular Characterization for Improved Sugarbeet Germplasm	J. McGrath (P)	7/1/03
	Improved Food Quality in Dry Bean Using Genetic and Molecular Approaches	R. Lu	11/28/03
Minnesota			
St. Paul			
	Wild Rice Breeding and Germplasm Improvement	C. Vance (P)	9/1/03
	Genetic and Genomic Approaches to Improve Disease Resistance and Nutrient Value in Wheat	D. Garvin (P) C. Vance	6/26/03
	Genomics, Germplasm Evaluation, and Genetic Improvement of Oats	H. Rines (P)	6/11/03
Mississippi			
Stoneville			
	Cropping Systems Research	L. Young (P) E. Walker E. King	2/25/04
	Combining Molecular and Conventional Techniques to Improve Cotton Fiber Yields and Quality	J. Scheffler (P) E. Taliercio R. Turley L. Young	9/27/03
	National Cotton Variety Test Program	W. Meredith, Jr. (P)	7/27/03
	Develop Soybean Genotypes and Management Systems for Early Season and Stress Environments	R. Paris (P) L. Young N. Bellaloui C. Koger	7/4/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Genetic-Physiological Team Research to Improve Production, Fiber Quality and Competitive Ability of Cotton	W. Meredith, Jr. (P) W. Pettigrew L. Young J. Johnson L. Zeng	10/1/03
	Genetic and Cultural Methods to Reduce Soybean Yield Losses to Diseases and Environmental Stress	J. Ray (P) J. Smith A. Mengistu L. Young	7/4/03
	Cultural and Genetic Methods to Manage Reniform Nematode in Cotton	L. Young (P) S. Stetina E. Sachs	2/11/03
	Alternative Crops and Value-Added Products for Mississippi	L. Young (P)	12/15/04
	Resistance Monitoring and Resistance Management of Lepidoptern spp. Infesting Bt Cotton	J. Adamczyk (P) G. Snodgrass C. Blanco O. Perera J. Gore	10/1/00
Poplarville			
	Vegetable and Ornamental Research in the Gulf South	J. Spiers (P) D. Boyd W. Copes H. Sakhanokho T. Rinehart G. Fain	9/5/03
	Small Fruit Cultural and Genetic Research for the Mid-South	J. Spiers (P) S. Stringer B. Smith	3/26/04
Mississippi State			
	Enhancing Corn with Resistance to Aflatoxin Contamination and Insect Damage	W. Williams (P) T. Brooks L. Hawkins G. Windham M. Clements	5/31/03
	Germplasm Enhancement and Genetic Improvement of Cotton	J. Jenkins (P) S. Saha J. McCarty, Jr.	8/17/03
Missouri			

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
Columbia			
	Breeding and Molecular Genetics of Corn	P. Beuselinck	11/16/03
	Genomic Characterization and Manipulation of Aluminum Tolerance and Other Value-Added Traits in Wheat	J. Gustafson (P)	6/18/03
	Plant Resistance, Biology, and Resistance Management of Insect Pests of Corn	B. Hibbard (P)	6/3/05
Nebraska			
Lincoln			
	Genetic Improvement of Sorghum for Enhancing Energy Yield, Nutrient Availability, and Disease Resistance	J. Pedersen (P) D. Funnell	4/11/03
	Genetic Improvement and Evaluation of Hard Winter and Spring Wheats	R. Graybosch (P)	5/31/03
New York			
Ithaca			
	Evaluation of Pest, Disease Resistance, and Stress Tolerance in Grape and Apple Rootstocks	P. Cousins (P) G. Fazio C. Owens A. Baldo A. Garris L. Cadle Davidson	4/24/03
	Genomic Approaches to Improving Transport and Detoxification of Selected Mineral Elements in Crop Plants	L. Kochian (P) T. Thanhouser	5/11/05
North Carolina			
Raleigh			
	Enhancing the Genetic Base of Corn with Genomics and Breeding	J. Holland (P) P. Balint-Kurti S. Szalma	11/2/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Increasing the Competitive Position of U.S. Soybeans in Global Markets through Genetic Diversity and Plant Breeding	T. Carter (P) R. Upchurch P. Bishop J. Burton D. Israel P. Kwanyuen	6/16/04
	GEM (Germplasm Enhancement of Maize) - Raleigh	P. Balint-Kurti (P)	10/1/04
	Fundamental Mechanisms for Genetic Alteration of Soybean Quality and Productivity	D. Israel (P) P. Kwanyuen J. Burton T. Carter R. Upchurch	7/26/02
North Dakota			
Fargo			
	Enhancement of Sunflower Germplasm Diversity through the Use of Wild Species and Genome Characterization	B. Vick (P) C. Jan G. Seiler J. Hu	6/4/03
	Enhancing Genetic Diversity, Processing Quality, and Root Maggot Resistance in Sugarbeet	L. Campbell (P)	7/16/03
	Development of Genetically Diverse, Pest Resistant Sunflower Germplasm	J. Miller (P) T. Gulya L. Charlet	5/29/03
	Genomic Relationships and Cytogenetic Manipulation of Durum Wheat by Classical and Molecular Techniques	P. Jauhar (P)	7/24/03
	Using Genomics and Genetics to Enhance disease Resistance and Quality in Hard Red Spring and Durum Wheat	J. Faris (P) S. Xu S. Chao Y. Tai	6/3/04
	Genetic Improvement of Barley	L. Dahleen (P)	3/20/01
	Oat Quality Improvement	D. Doehlert (P)	9/12/04
	Insect Genomic Biodiversity and Molecular Regulation of Diapause	R. Roehrdanz (P) G. Yocum	5/25/05
	Development Of Cold Storage Technology For Mass-Reared and Laboratory-Colonized Insects	R. Leopold (P)	5/25/05

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Enhance Hard Spring and Durum Wheat Quality and Utilization	G. Hareland (P) L. Grant	9/10/04
	Sucrose Accumulation and Retention in Sugarbeets	K. Klotz (P)	3/27/01
Ohio			
Wooster			
	Germplasm Improvement and Virus Diseases of Soybean	R. Mian (P) R. Gingery M. Redinbaugh	10/01/00
Oklahoma			
Woodward			
	Germplasm Development for Southern Plains Agricultural and Rangeland Ecosystems	T. Springer (P) R. Gillen P. Sims J. Goldman	9/4/02
Stillwater			
	Genetic Improvement of Insect Pest Resistance in Wheat, Barley, and Sorghum	D. Porter (P) Y. Huang D. Mornhinweg	6/11/03
	Biologically Based Cereal Aphid Management	N. Elliot (P) J. Burd G. Puterka K. Shufran	4/7/05
	Improvement of Disease Resistance and the Quality of Peanut	H. Melouk (P) K. Chenault	2/7/03
Lane			
	Physiological and Genetic Basis of Postharvest Quality and Phytonutrient Content of Fruits and Vegetables	P. Veazie_Perkins (P) A. Davis W. Fish	6/16/04
Oregon			
Corvallis			
	Genetics, Genomics and Germplasm Development of Hops	J. Henning (P) G. Banowetz D. Gent	3/10/04

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Physiology, Biochemistry, and Genetic Improvement of Small Fruit Crops	D. Bryla (P) C. Finn R. Doss J. Loper	7/22/04
	Production Systems to Promote Yield and Quality of Grapes in the Pacific Northwest	J. Tarara (P) K. Shellie J. Loper J. Lee R. Martin	11/16/03
Puerto Rico			
Mayaguez			
	Characterization, Conversion, and Improvement of Common Bean Germplasm	T. Porch (P)	5/28/04
	Development of Integrated Systems for Subtropical/Tropical Fruit Crop Production	R. Goenaga (P) D. Jenkins	10/1/02
South Carolina			
Charleston			
	Evaluation and Genetic Enhancement of Cole Crop and Cucurbit Germplasm	M. Farnham (P) A. Levi	7/27/03
	Genetic Improvement of Sweetpotato and Snap Bean for Multiple Pest Resistance and New Uses	J. Bohac (P)	3/27/04
	Genetic Improvement of Southernpeas and Peppers	R. Fery (P)	9/10/03
Florence			
	Enhancing the Sustainability of Cotton Production in the Southeast USA	P. Bauer (P) J. Novak B. Campbell	6/5/04
Texas			
College Station			
	Genetics and Improvement of Pecan	T. Thompson (P) L. Grauke	3/4/03
	Sorghum Genomics and Germplasm Evaluation	R. Klein (P)	4/17/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Conservation, Evaluation and Genomic Characterization of Cotton Genetic Resources and Associated Information	R. Kohel (P) J. Yu L. Hinze	6/21/03
	Genetic Enhancement of Warm-Season Perennial Forage and Range Grasses	B. Burson (P)	9/25/02
Beaumont			
	Application of Rice Genomics to Develop Sustainable Cropping Systems for the Gulf Cost	A. McClung (P) R. Fjellstrom S. Pinson	7/11/03
Lubbock			
	Molecular and Genetic Enhancement of Drought and Temperature Stress Tolerance in Sorghum	Z. Xin (P) J. Burke C. Franks G. Burow	6/10/04
Utah			
Logan			
	Improved Plant Genetic Resources for Pastures and Rangelands in the Temperate Semiarid Regions of the Western U.S.	J. Chatterton (P) T. Manco M. Peel S. Larson B. Waldron K. Jensen T. Jones R. Wang D. Johnson B. Bushman J. Robins	10/16/02
Washington			
Pullman			
	Germplasm Enhancement, Genetics and Disease Management of Cool Season Food Legumes	F. Muehlbauer (P) K. McPhee W. Chen	7/1/03
	Identification and Deployment of Genes to Reduce Production Risks and Improve Quality in Club and Soft White Winter Wheat	C. Steber (P) C. Garland D. Skinner	6/18/03

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Characterization of Expression Patterns of Stress Tolerance Genes in Wheat	D. Skinner (P)	6/18/03
	Enhance Wheat Quality and Utilization in the Western U.S.	C. Morris (P) D. Skinner	9/21/04
Prosser			
	Evaluation of Advanced Potato Breeding Lines for Biotic and Abiotic Resistances and Yield	C. Brown (P)	10/01/01
	Bean Germplasm Enhancement and Improved Disease Management of Edible Legumes	P. Miklas (P) A. Alva L. Porter	7/16/03
	Potato Variety Improvement through Gene Transfer and Virological Studies	C. Brown (P) D. Navarre A. Alva J. Crosslin	7/16/03
Washington, DC			
	Genetic Improvement of Landscape Trees for Superior Pest Resistance	J. Hammond	11/19/03
	Genetic Improvement of Floral Crops for Pest Disease, and Stress Tolerance and Ornamental Qualities	R. Griesbach (P) J. Hammond R. Jordan	12/31/03
	Genetics, Genetic Resource Evaluation, and Genetic Improvement of Landscape Trees and Shrubs	M. Pooler (P)	11/23/03
	Genetics, Genetic Improvement and Improved Production Efficiency of Nursery Crops	S. Reed (P) D. Fare	11/19/03
West Virginia			
Kearneysville			

Location	Project Title	Scientists (P=Principal Investigator)	Start Date
	Genetic Improvement of Fruit Crops	R. Scorza (P) C. Srinivasan R. Bell A. Callahan D. Liu D. Swietlik C. Dardick	4/30/03
Wisconsin			
Madison			
	Potato Genetics, Cytogenetics, Disease Resistance, and Prebreeding Utilizing Wild and Cultivated Species	S. Jansky (P) P. Simon D. Halterman	4/24/03
	Allium, Cucumis, and Daucus Gemplasm Enhancement, Genetics, and Biochemistry	P. Simon (P) M. Havey J. Staub	7/3/03

Component III

Location	Project Title	Scientists (P)=Principle Investigator	Start Date
California			
Albany			
	Database and Bioinformatic Resources for Small Grains Research and Crop Improvement	O. Anderson (P) Y. Gu V. Carollo G. Lazo D. Matthews	7/24/03
	Plant/Crop Genome Sequencing	S. Hake	4/16/03
Iowa			
Ames			
	Curation and Development of Soybase and Its Integration with Other Plant Genome Databases	R. Shoemaker (P) L. Lewis	3/21/03
	Database of Maize Genome Information	M. Scott (P)	4/3/04
Mississippi			
Stoneville			

Location	Project Title	Scientists (P)=Principle Investigator	Start Date
	Genomics and Bioinformatics Research in Catfish, Cotton, and Soybeans	B. Scheffler (P) G. Waldbieser	10/1/02
Missouri			
Columbia			
	Maize Genome Database	M. Polacco (P)	10/01/04
New York			
Ithaca			
	Dissecting Complex Traits in Maize by Applying Genomics, Bioinformatics, and Genetic Resources	E. Buckler (P) P. Bradbury	6/03/05
	Comparative Plant Genomics	D. Ware (P)	8/24/03