NCRPIS Annual Report - 2005
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NORTH CENTRAL REGIONAL PLANT INTRODUCTION STATION
NC-7 ANNUAL REPORT, JANUARY 1 - DECEMBER 31, 2004

I. PROJECT TITLE:

NC-7 "Plant Germplasm and Information Management and Utilization."

II. COOPERATING AGENCIES AND PRINCIPAL LEADERS (current):

A. Administrative Advisor

*W. Wintersteen, Iowa

B. Regional Coordinator

*C. Gardner, ARS, Iowa

C. State Experiment Stations Representatives

1.	Illinois	*T. Hymowitz	7. Missouri	*P. Beuselinck
2.	Indiana	*J. Janick	8. Nebraska	*D. Baltensperger
3.	Iowa	*C. Brummer	9. N. Dakota	*B. Johnson
4.	Kansas	*M. Stamm	10. Ohio	*D. Francis
5.	Michigan	*A. Iezzoni	11. S. Dakota	*A. Boe
6.	Minnesota	*S. Hokanson	12. Wisconsin	*W. Tracy

*Voting members

D. U. S. Department of Agriculture

1. ARS National Program Staff, Plant Germplasm	*P. Bretting
2. ARS Plant Exchange Office	*E. Garvey
3. ARS Area Director, Midwest Area	S. Shafer
4. Cooperative State Research, Education and Extension Service	A. Thro
5. Natural Resources Conservation Service	*Vacant
6. National Center for Agric. Util. Research	*T. Isbell
7. National Center for Genetic Resources Preservation	*H. Shands

E. North Central Regional Plant Introduction Station, Ames, Iowa

See organizational chart, Figure 1 in the Appendix.

III. PROGRESS OF WORK AND PRINCIPAL ACCOMPLISHMENTS:

Our mission continues to focus on acquisition, documentation, regeneration, characterization, evaluation, and distribution of germplasm and associated information. The world's biological diversity and plant genetic resources (PGR) continue to be threatened by climatic, social, political and environmental pressures. Demand for genetic diversity and development of supporting technologies to increase the availability of PGR continues to increase.

The quality of collections, associated information, and service provided to the research and educational communities continues to build upon the work of generations of plant scientists,

implementing new technologies and new learning. Quality information delivery systems are critical to successful use of plant genetic resources and realizing the potential they offer.

Available resources (approximately $2.2 M from ARS and $0.53 M from NC7 funds) were devoted to this mission, in order to secure and maintain our plant genetic resources, staff, support equipment, laboratory and field activities, and research designed to provide information which improves the quality, value and utility of the collections.

Progress on updating the Operations Manual is nearing completion, but the document must be dynamic in order to reflect improved practices based on new technologies and learning. The Operations Manual provides relevant reference and/or training materials to guide our staff in their curatorial operations and procedures, ensuring high standards and quality of operations. NCRPIS Staff contributed to the revision of the Acquisition and Distribution Policies for the NPGS, and to the Active Site section of the revised NPGS Operations Manual.

Personnel changes - June, 2005 – May, 2006:
Departures:
Stacey Winter, USDA-ARS Research Unit Secretary, October, 2005.
Cris Nass, USDA-ARS Research Office Automation Asst., October, 2005.
Josh Davis, USDA-ARS LA Computer Intern, October, 2005.

New Hires:
Marci Bushman, USDA-ARS Research Unit Secretary, December, 2005.
Fred Engstrom, USDA-ARS Agri. Res. Science Technician, GEM Project, August, 2005.
Matt Lively, USDA-ARS Agri. Res. Science Technician, Maize Curation, November, 2005.
David Losure, Iowa State University Agricultural Specialist I, Maize Curation, January, 2006.
Philip Biggs, USDA-ARS Computer Intern, December, 2005.
Robert Frey, USDA-ARS Computer Intern, June, 2005.

Transitions:
Mark Millard, from ISU Maize Curator to USDA-ARS Maize Geneticist, October, 2005.
Andy Smelser, from Term to Permanent USDA-ARS Term Agri. Res. Science Technician, GEM Project, April, 2006.

Visiting Scientist:
Ana Gulbani, Academy of Agricultural Sciences of Georgia, Tbilisi, Republic of Georgia, arrived on October 31, 2004, and departed in December, 2005. Ms. Gulbani learned all phases of germplasm management and curatorial activities, and focused on maize curatorial activities during the last six months of her time in Ames.

Management of Federal STEP (Student Temporary Employees):
Approximately 60 ISU students were hired to fill 26 FTE positions which supported curatorial projects, viability testing, IT support and development, and farm and facilities operation. Students were interviewed and selected by ISU Program Manager Larry Lockhart or ARS IT Specialist Peter Cyr. Cris Nass and Stacey Winter, followed by Rhonda Bartlett and Marci Bushman, managed all hiring processes, with support and guidance by Ames ARS HR Specialist Lynnette Richey.

Construction and Facilities:
Highlights include completion of construction of a new machine shed; reshaping and seeding of two primary waterways on the Plant Introduction Farm; removing valve pits and hydrants from the former swine farm area to facilitate plot work; completion of remodeling of the maintenance shop and headquarters building restrooms; software upgrades for and expansion and maintenance of the

keycard access security system. The neighboring Organic Student Farm is being relocated; the land involved will be made available by the ISU Agricultural Experiment Station for NCRPIS use.

Design was completed for an environmental control system which will be used to monitor and control greenhouse, seed storage and dryer conditions. Greenhouse facilities are needed, but we lack funding to pursue development at this time. Expansion of the headquarters building is needed, but is not a high priority.

Equipment:
New equipment acquisitions in 2005 included a new PCR machine, a Zeiss microscope with digital photography capabilities for the pathologist, 176 new 7x7x20' regeneration cage frames and screens, a row mulch wagon, cages for cooperative *Daucus* increases, a replacement compressor for seed storage environmental control, and a gently used New Holland twin-rotor combine which was modified for two-plot maize research harvest.

Seventeen new PC workstations were deployed to permanent staff based on prioritized needs; the backup server was upgraded; new server hardware was installed for accession image storage; the network security firewall was upgraded; a new email server and Microsoft Exchange 2003 were installed. See Computers and Telecommunications Section for software upgrade details.

IV. PROGRESS IN GERMPLASM AND INFORMATION MANAGEMENT, RESEARCH, AND EDUCATION (C.A. GARDNER):

(Part IV. summarizes the accomplishments and progress presented in greater detail in the individual staff reports in the document.)

Acquisition highlights:
Total acquisitions in 2005 numbered 282 accessions, down from 450 in 2004. These included 75 maize accessions, of which 34 were expired PVP lines, 21 were U.S. Crop Science Registered (CSR) lines, and the remainder were CSR lines from Nigeria. A black seeded grain amaranth cultivar was acquired from Lithuania. Fifteen new landrace type umbels of anise, coriander and cumin were received from the Palestinian Territory. Eighty one accessions of ornamentals and mints were acquired from the Republic of Georgia and the Palestinian Territories. The medicinal plants collection grew with the addition of 59 new accessions, with emphasis on native *Hypericum* and Federally endangered *Echinacea laevigata*. Seven new *Cucumis* accessions were received and documented.

The Sunflower CGC's goal of acquiring accessions representative of the complete geographic distribution of all 66 *Helianthus* taxa was achieved. Collection trips by Curator Marek and various cooperators yielded cultivated *H. annuus*, *H. niveus ssp tephrodes*, *H. niveus ssp canescens*, *H. cusickii*, *H. deserticola*, *H. pumilus*, *H. nuttallii ssp nuttalli*, and *H. carnosus*.

Seven *Brassica napus* accessions originating from Sweden were transferred to the NCRPIS from the NCGRP following expiration of their PVP certificates.

See Table 1 for additional details.

Regeneration and Maintenance Highlights:
Over 1430 total accessions were grown for regeneration and 989 harvested. Approximately 1,412 accessions were made available to the public. Over 4,120 accessions were tested for viability, approximately 9% of our holdings. About 840 accessions were backed up at the NCGRP; 77% of our holdings are currently backed up, and 73% are available. This represents an increase of 1% of the accessions that are backed up and 2% that are available.

The success of cooperative *Helianthus* regeneration efforts with the National Arid Land Genetic Resources Unit in Parlier, CA was greatly enhanced by a protocol change. The NCRPIS oilseeds staff now germinate and ship established seedlings to Maria Jenderek; good survival rates and good work on the part of Parlier have resulted in successful harvests. In Ames, substantial progress was made on establishing wild perennial populations; this is partly due to more focus applied to increasing their availability and partly due to enhancements made to germination and seedling establishment methods.

Hypericum regeneration was challenged by the first identification of an infestation by *Colletotrichum gloeosporioides*, a devastating stem disease that cases rapid death of *Hypericum*. Curator McCoy and Pathologist Block are closely monitoring greenhouse and field plantings, and are learning about to manage these diseases.

The Vegetable Project utilized large (5x5x40') cages for *Cucurbita* regenerations. *Cucumis melo* regenerations were enhanced by use of an isolation system developed by NCRPIS staff members that prevents splashing droplets from adjacent trays of seedlings in the greenhouse via use of a bottom-watering system. No bacterial fruit blotch was found on the melon seedlings grown using this system in the greenhouse or later in the field post-transplanting, eliminating the need to treat seeds with hydrochloric acid solution during processing.

Maize regeneration efforts in Ames were reduced in 2005 due to short staffing. Regeneration efforts in Puerto Rico were increased. The only accession of *Zea nicaraguensis* in the NPGS was successful regenerated, a result of two years of greenhouse propagation effort by Curator Millard's team and the help of visiting Republic of Georgia scientist Ana Gulbani. Short-term, collection availability will decrease because the rate of growth exceeds the rate of regeneration.

The Horticulture Project expanded a three-year cage field established for shrubs, annuals and tall Malvaceae in 2004, and also established a new, three-year cage field focusing on *Potentilla*, Lamiaceae, and Malvaceae in 2005. All horticultural plantings are in extremely good condition, due to the energies of technician Jeff Carstens and Mark Widrlechner's direction.

Curator Brenner utilized a long, warm-humid summer dormancy breaking method which resulted in improved germination of wild *Spinacia tetrandra*. Further research is needed to determine if this method is broadly applicable for wild spinach germplasm and perhaps other species with seed dormancy issues.

See Table 2 for additional details.

Distribution:
2005 distributions included 22,300 seed packets or plants in 915 orders to external germplasm requestors (Table 3A). These represented 12,289 accessions, or 25% of the collection, consistent with a historical average of 20-25% annual distribution of all holdings. Approximately 67% of these were to U.S. and 33% to international requestors, also consistent with historical averages. Additionally, 10,428 items were distributed within the NCRPIS (Table 3B). Order information by crop is presented in Table 5. CSREES regional distribution history is presented in Table 6.

Seed order numbers have increased 18% over 2004 levels, primarily due to interest in expired maize PVP lines that are newly available (see Maize Curation project report for specific details) and requests from home gardeners for many popular species and cultivars, a large request from Embrapa-Cenargen, Brazil for 1175 accessions of cultivated and wild *Helianthus*, and Brassica for disease resistance and breeding research. The number of accessions distributed increased by nearly 15%. Demand for *Cucumis* and *Cucurbita* continued to increase. Although our resources cannot

support maintaining and distributing the collections to home gardeners, we have tried to use this development to educate the public about plant genetic resource conservation and encourage interested individuals to save seeds, conserve and share germplasm and associated information, and to pursue their own research interests with the materials. Home gardeners are redirected to other sources of commercially available materials.

We distributed 208 plants of five accessions to 18 sites for evaluation in the Regional NC-7 woody ornamental trial evaluations across the Midwestern U.S, with an additional 77 plants of these accessions provided to 11 public gardens. In the process, M. Widrlechner and J. Carstens met with cooperators in South Dakota, North Dakota, and Minnesota. Clonally propagated materials in demand include *Salix* and *Fraxinus*.

Leaf samples of *Hypericum* and *Echinacea* were distributed for molecular-marker analysis to Jonathan Wendel and Eve Wurtele of Iowa State University.

Efforts were initiated to modernize our Accession Performance Reporting system (APR) in order to link the purpose and outcomes of research efforts with specific orders and accessions electronically and enable web-based capture of responses to a database. Currently, APR reports are hard copies kept in files which do not lend themselves to analysis of short-term or long-term impacts of use of germplasm. A prototype will be presented at the RTAC meeting.

Evaluation and Characterization:
Over 2,040 accessions were planted for observations and characterization for a wide array of descriptor information; over 41,727 observations were transferred to the GRIN database (http://www.ars-grin.gov/npgs/) in 2005 on a total of 10,491 accessions (Table 4). The vegetable, maize and medicinal plant projects contributed the bulk of the descriptive data entered in 2005. An extensive collection of evaluation data received from NC-7 trial-site cooperators was loaded into a local Access database and prepared for posting on the Internet.

AFLP analyses for use in genetic diversity pattern study, and fatty acid compositional analyses of coriander were completed by graduate student Pedro Lopez in cooperation with Terry Isbell, NCAUR, Peoria, IL. The information will be analyzed together with previously collected morphological, phenotypic and biochemical traits to establish phylogenetic associations. Mr. Lopez is currently completing his dissertation.

Graduate student Von Mark Cruz completed morphological and flowering data characterization of the *Brassica napus* collection, extraction of DNA, sequenced introns of flowering gene loci Flc 1 and 3. He completed a project to assess SSR profiles of bulked *B. napus* accessions, and incorporated morphological data into the analyses to determine the appropriateness of bulking accessions. He also conducted a study to determine whether caged regeneration methods for *B. napus* preclude pollen flow from cage to cage and ensure that the original genetic profile is preserved. Mr. Cruz is currently completing his dissertation.

DNA capture on Whatman FTA™ paper storage media was utilized by Laura Marek to capture DNA for tissue analysis from the wild populations collected in 2005. This method was also been used to capture *Brassica* DNA by graduate student Von Mark Cruz and by C. Gardner, Lindsay Werth and Deb Muenchrath for the Southwestern U.S. Maize characterization project.

A cooperative project with Mike Owen of Agronomy and Mark Dahmer of BASF focused on screening wild *H. annuus* and *H. petiolaris* species for resistance to the imidazalinone imazamox herbicide Beyond®. This effort was part of BASF's efforts to satisfy requirements to allow the marketing of Clearfield™ sunflowers in Canada.

A cooperative internal effort to develop a system to control *Systole* chalcid insect infestation in *Coriandrum* seeds was successfully completed.

Information technology and telecommunications:
Digital imaging efforts were greatly enhanced by implementation of software developed by Pete Cyr and Mark Millard to automate the capture and loading of digital images. Over 4,240 images were captured by the curatorial teams in 2005; they will be rapidly transferred to the GRINdatabase in 2006 (Table 4).

The MaizeGDB (http://www.maizegdb.org/) now links molecular genetic information to accession information located in the GRIN database; its order module enables genomic researchers to order maize accessions through GRIN. Efforts have been initiated to better link phenotypic information held in GRIN with genomic information held in MaizeGDB.

The digital compendium of the Races of Maize Volumes was completed in 2005 with the capture of the Spanish version of the Races of Maize in Mexico, 'Raza de Maiz en México.'

Dave Kovach created a new Internet site for the Station and for the Ames Area USDA Civil Right advisory Committee's website, continued efforts on software forms development.

See full curatorial team reports for more detail.

Testing germplasm's germination, viability, and health:
Nine percent of the NCRPIS holdings, or 4,124 accessions, were tested for viability in 2005; this included 7% of the maize collection, 14% of Curator Brenner's collections, 13% of the vegetable collection, and 24% of the medicinal plants collection (Table 2).

The viability testing group also investigated new methods to promote germination of dormant seeds. *Daucus* seeds were dissected to evaluate variation in the degree of embryo development. *Acer* seeds were tested to determine whether embryo excision would promote germination.

David Brenner investigated a long, warm-humid summer dormancy breaking method which resulted in improved germination of wild *Spinacia tetrandra*. Further research is needed to determine if this method is broadly applicable for wild spinach germplasm and perhaps other species with seed dormancy issues.

Field observations were made for sunflowers resistance reactions to Septoria leaf blight disease, maize for artificially infested Stewart's wilt, vegetables for viruses and bacterial diseases, naturally occurring disease of *Brassica,* and downy mildew of sunflower. Monitoring for naturally occurring pathogens to preclude accidental distribution of infested seed is an ongoing part of germplasm maintenance and distribution activities. A greenhouse study of within-in plant movement of squash mosaic virus showed that the virus moved from an inoculated leaf near the center of the plant to the growing tips within 7 to 10 days, which reinforces the importance of using screened cages in the field to exclude beetle vectors of SqMV.

The first occurrence of anthracnose on *Hypericum* plants was observed at the Plant Introduction farm.

Pathologist Charles Block in cooperation with ISU Seed Science's A. Fessehaie and L. Shepperd developed a real-time PCR assay for detection of *Pantoea stewartii* from maize seed, and are currently evaluating the sensitivity and reproducibility of the assay for detecting the pathogen from infected seeds.

Insect management:
Our entomologists continue to develop thoughtful insights which help better manage and utilize alfalfa leaf cutter (ALC) bees in our regeneration programs. ALCs have proven most useful with plants with small to medium size flowers of a flat nature, and warm (26 C or 80 F or above), mostly sunny, dry growing conditions. ALCs seem unwilling to compete with other pollinator insects for pollen or nectar sources. These bees are preferred by our staff because they do not sting and require less management on the part of Entomology staff members. Please see Entomology research and service report sections for a great deal of fascinating, detailed information.

Protocols and recommendations on the use of ALC pollinators for caged clover pollinations were provided to Healthcliffe Riddell of the University of Wisconsin.

A commercial 'graftless' queen rearing technique was tested in 2005. This technique forced the queen to lay eggs in a selected number of cells which were then placed into a queenless hive so the colony would finish caring for the queen cells. After 12 days, cells were removed and placed in a queenless nucleus hive to start new colonies. This technique worked very well, but is not productive enough for our applications.

A pollinator 'tip of the month' feature was developed and communicated to staff members to help them educate their support staff on how to understand insect behavior, perform plant-related tasks such as watering and chemical applications so that the least harm is done to the pollinator insects.

Use of alfalfa leaf cutter bees (ALC) for sunflower, Brassica and vegetable curation programs was evaluated in the field and greenhouse in 2004-2005, and for some ornamental species in the field in 2005. Seed production using ALC's is being evaluated to compare their efficacy with honeybee, fly and bumblebee use in seed regeneration efforts. More extensive use of non-stinging, solitary ALC bees could improve worker safety and decrease program costs. In-house fly rearing was terminated in 2004; fly pupae are now purchased, resulting in cost-savings and enabling our researchers to spend their time more effectively.

Enhancement:
Curator Brenner released two ornamental amaranth varieties through Iowa State University, Pillar Orange and Pillar Red. These varieties have better lodging resistance than older ornamental cultivars and non-shattering seed cases which improve performance for cut-flower use. Brenner continues to cooperate with the Univ. of Nebraska's David Baltensperger to develop new proso millet cultivars that are genetically broadly based.

The Germplasm Enhancement of Maize (GEM) Project and its public collaborators released 17 lines in 2005; a total of 135 lines have been released since 2001 that resulted in the introgression of exotic maize germplasm into elite domestic lines. These lines were released because they possess unique genetic diversity with good yield agronomic performance; some have unique grain quality or compositional attributes for enhanced feed, food or nutritional use. Others have disease or insect resistance attributes, or unique characteristics for improved silage quality.

Outreach and Scholarship:
Over 300 visitors toured the NCRPIS during 2005. Our staff participated in teaching students from the grade K to postgraduate level, provided outreach events to civic and other organizations about germplasm conservation and management, and the work done at the NCRPIS. Scientific and technical staff members continue to publish scholarly journal articles, make presentations at scientific meetings, and supervise graduate research programs.

Current and future foci:
Horiculturist M. Widrlechner serves as chair of a national Technical Review Team that provides technical direction and oversight to an ARS project to update the USDA Plant Hardiness Zone Map using the best available technologies and data sets, and make it accessible via the Internet. Project completion is anticipated in 2006.

Efforts to characterize the Southwestern U.S. maize landraces continue; completion of this project is anticipated in 2007.

Real-time PCR analytic methods will continue to be evaluated and modified by Pathologist Block's team for routine detection and identification of multiple pathogens from seeds. Efforts continue to identify vectors of cucurbit viruses that may infest plants through screen cages.

The anticipated hiring of a second germplasm program assistant in 2006 will assist in efforts to improve passport and provenance information, and enable our curators to accelerate the process of assigning PI numbers to Ames numbered accessions, in addition to their normal duties.

Software development efforts will be pursued to help the Entomology Team and curators manage the production and introduction of pollinator insects into cages in a timely, efficient manner. Our Accession Performance Reporting effort is undergoing made revision and will be made available to requestors via a web-delivery format. This is part of an effort to improve our ability to capture and assess the impact of use of germplasm. This information may be important in determining the focus and structure of germplasm conservation and utilization programs in the future.

Software development to support data capture on handheld PC's will continue in 2006-2007. Use of servers and software to manage digital image and document image files will be implemented.

V. IMPACTS OF GERMPLASM USE BY NORTH CENTRAL REGIONAL RESEARCHERS:

Impacts of germplasm use by the researchers at the NCR institutions:
A detailed list of examples of germplasm use in research being conducted at NCR institutions was not requested of the RTAC members this year. Please see Table 6 for a summary of the various CSREES regions order history. Requests for germplasm continue to increase for research as well as non-research use. Requests become increasingly better targeted as the quantity and quality of information associated with the collection improves.

Eleven of the 12 NCR states participate in the NC-7 woody ornamental trials, which marked their 50th anniversary in 2004. These trials represent the longest running evaluation trials in the U.S.; they are a key component for identification of useful materials for the ornamentals industry which have the necessary traits for adaptation in our cold, arid winters, and highly variable summers. This program effectively links the institutions of the NCR and beyond. An improved Accession Performance Reporting Process will enable better assessment of these linkages in the future.

The linkage of the GEM Project, the maize curation project, and public and private collaborators throughout the U.S. is resulting in synergy which is facilitating the use of exotic maize germplasm by public and private sector maize researchers. This unique partnership offers great potential for diversifying the genetic base of U.S. maize production, the purpose of the GEM Project.

Linkages among project participants and with other projects/agencies and contributions of the Regional Technical Advisory Committee:
Linkages are driven primarily by common research interests and objectives and by the heritage of the germplasm material utilized for research and education. All states utilize germplasm provided by the NCRPIS and many of the other 20 sites involved in the NPGS; the states have a complex array of collaborative research efforts between their institutions, and with the plant genetic resource curators at the NPGS sites.

The regional technical advisory committee (RTAC) has provided valuable direction in the following areas:

- requesting and suggesting organizational structure of information needed to determine project impact and provide accountability. This includes advice on useful formats for analyzing and evaluating the nature of distributions, who they benefit, and how benefits are realized, which are essential for determining the impact and value of the project
- identifying needed improvements to the public GRIN interface
- providing input from their respective AES Directors to curators, genebank and other administrators
- providing guidance to increase the NCRPIS program's relevance to NCR stakeholders
- providing technical expertise, particularly in the areas of diversity assessment and taxonomy
- providing added breadth in understanding issues at genebanks beyond the NCRPIS
- understanding challenges faced by public researchers partnering with other public institutions' researchers, both governmental and non-governmental. This has provided useful insights for ARS and NCR administrators to guide programmatic decision-making, as well as operational guidance; this function is key because of its direct impact on the public interest as well as the specific research interests of more directly involved stakeholders.

The technical committee gatherings provide an opportunity for the AES Directors' representatives to learn about and understand strategic issues which impact how their institutions operate and how they can cooperate more effectively to address their mission in today's environment, and then provide this information to their Directors.

Some of the NC-7 RTAC's specific suggestions and contributions from their 2005 Annual Meeting include the following:

- Crop Germplasm Committees must be encouraged by the leader of the National Genetics Resource Lab and Data Management Unit to ensure that CGC annual reports are made current and that they fulfill their obligations.
- Instructions should be developed and posted on the NPGS website on how to appropriately credit curators and other NPGS entities for their contributions to the success of research efforts.
- Creation of a distance education course for use of public GRIN be investigated, or the public interface be significantly improved for ease of use and information delivery
- Imaging methods used for herbarium specimens should be studied and implemented as appropriate by the NCRPIS.

VI. SUPPORT TEAM REPORTS:

A. Farm (L. Lockhart, L. Crim, B. Buzzell)

We supervised and coordinated daily operations at the NCRPIS farm, including management of all facilities, fields, and greenhouse space. We supervised or conducted pesticide applications in the field and campus greenhouses. We responded to maintenance requests from staff members at the farm and the campus location. We selected, coordinated and scheduled the student labor force of 26.0 FTE's. We coordinated and completed facility construction and upgrades.

Labor:
During 2005, 147 applications for hourly employment were received and reviewed. There were 51 interviews, resulting in 31 new hourly employees hired. Currently there are 47 Biological Science Aides (21.4 FTE) working at the NCRPIS.

NCRPIS FARM CREW Personnel:
Larry Lockhart (Program manager II) has been on staff since 1985.
Lloyd Crim (Equipment Operator III) joined the staff in March 1998.
Brian Buzzell (Farm Mechanic) joined the staff in May 2002.

Maintenance projects:
During the past year the farm staff initiated and completed several projects which enhanced the efficiency and safety of the station operations:

1) Coordinated the expansion of the Security Access System to the remodeled maintenance shop and new machine shed.
2) Finished remodeling of shop.
3) Coordinated construction of new machine storage shed.
4) Coordinated efforts with GEM, Research Leader and others to purchase and modify New Holland TR-88 combine for dual plot harvesting.
5) Installed new furniture in Unit Secretaries office.
6) Remodeled Horticulturalist office.
7) Finished remodeling and furniture installation in Germination Lab.
8) Reshaped and seeded two large waterways on the Plant Introduction Farm.

Purchasing:
Larry Lockhart coordinated purchasing for the NCRPIS farm: this task included gathering and summarizing requests, writing specifications, and obtaining supplies for the farm. Major purchases included the following:

1) One new vehicle.
2) New Holland combine.
3) 187 7x7x20 cage frames and covers.
4) Row Mulch Wagon
5) Replacement Compressor for Seed Storage
6) Cages for cooperative *Daucus* increases.

Tours:
This past year, we organized and conducted 18 tours. There were 301 visitors to the NCRPIS during 2005.

Staff Training:
We conducted two Tractor Safety training sessions and several Worker Protection Standard training sessions for the new student employees and existing staff.

Plans for 2006:

1) Prepare recently acquired ISU Student Organic Farm for incorporation in the PI operations.
2) Plan PI Headquarters expansion.
3) Manage installation of Computerized Greenhouse Control System.
4) Remove valve pits and hydrants from former swine farm to facilitate plot work in those areas.
5) Remodeling of restrooms in the headquarters building.

B. Information Technology and Telecommunications (P. Cyr)

Pete Cyr was assisted in IT duties by two part-time students. The following list outlines the progress made by the IT team during FY 2005 at NCRPIS.

Equipment:
As of December 2005, the NCRPIS has 71 workstations installed for use by permanent staff members and part-time temporary student help. There were 17 new workstations deployed in 2005 to permanent staff based on prioritized needs. Where possible, the displaced computers were re-commissioned for light duty work in other areas of NCRPIS and/or donated to local community school systems.

Due in large part to increased volumes of accession images captured at NCRPIS, the backup server was upgraded to increase throughput during the nightly backup window.

Upgraded the network security firewall and created 17 new VPN security policies to enable computers located outside of the Farm network to access the servers and email system on the Farm site.

Installed new server hardware for email and installed Microsoft Exchange 2003.

Installed new server hardware for storage of accession images and enabled FTP on this server for use in mass loading of archived images.

Software:
All of the workstations at NCRPIS are standardized on Windows XP with Service Pack 2 installed for increased security and reliability. Daily updates to anti-virus and periodic updates to anti spy-ware definitions help to ensure that these workstations stay healthy and productive.

During 2005, Altiris™ remote computer management software was removed from all workstations at the NCRPIS. The Altiris™ software was replaced with PatchLink™ software for the installation of security patch updates. PatchLink™ software is the standardized solution for patch management in the USDA-ARS.

In 2005, NCRPIS upgraded several administrative workstations with standard USDA/ARS software packages including ARIS, CATS and PCMS.

Upgraded and maintained a new version of Symantec Anti-Virus Corporate Edition to facilitate virus protection on all of the servers and workstations at the NCRPIS site.

Created and maintained a new software system to upload accession images to GRIN. This image loading software has greatly assisted the curators in removing backlogs of archived accession images that needed to be loaded to the GRIN system.

Enhanced the weather station software to make it easier to retrieve archived weather data. This weather data is used by curators for pollination and recording of observations during the growing season.

Maintained and enhanced the local intranet website used by NCRPIS for dissemination of group information. Assisted D. Kovach in development and maintenance of web pages located on the consolidated ARS website.

Documentation:
Supplemented the Races of Maize (an electronic compendium of important Maize literature) with a Spanish volume version, the "Raza de Maiz en México". Oversaw the production of 70 copies of the 5 CD disc-set that were distributed to members of the Maize CGC during the 2005 meeting in Chicago.

Posted committee meeting minutes and other farm operation information on the NCRPIS website.

Created and updated Disaster Recovery Documents for use during recovery of individual server systems at the NCRPIS Farm.

Plans for 2006:
Continue to replace NCRPIS workstations on an as needed basis (targeting a 3-5 year lifespan for daily use workstations).

Migrate all NCRPIS email users to the new Microsoft Exchange 2003 email server and decommission the old Exchange 5.5 server.

Perform the annual upgrade of the Symantec anti-virus software used to protect the servers and workstations at NCRPIS.

Install new Help Desk system for tracking and organizing IT service and procurement requests.

Enhance the GRIN image loader software developed in 2005 to facilitate the use of this software at sites outside of NCRPIS.

Develop a DVD version of the Races of Maize compendium.

Work with CI3 (door-access security system vendor) to enable the NCRPIS sites security system to call out to a pager when an intrusion (forced entry) is detected at the Farm.

Develop, test and deploy applications to be installed and used on the Compaq iPaq PocketPCs. These applications will be designed to assist curators in collecting and analyzing field data. These field solutions will leverage the use of barcode technology wherever possible. The target date for initial use of these PDA based field solutions is the end of second quarter of 2006.

C. Seed Research and Computer Application Development (D. Kovach and M. Erickson)

Seed Research:
In 2005, D. Kovach collaborated on research with Sharon McClurg of the NCRPIS and Susan Stieve of the Ornamental Plant Germplasm Center (OPGC) to test the utility of x-ray digital imagery in estimating the degree of chalcid infestation in coriander seed. Coriander's two-seeded

fruit made the estimation difficult, but overall correlations between results obtained via image analysis and fruit dissection were significant. In another experiment with Sharon McClurg, we determined the effectiveness of chalcid control by LN2 treatment via seed dissections. We demonstrated that LN2 is an effective method to destroy chalcids in coriander seed. These results have been summarized in a paper submitted to Seed Science and Technology for review.

In 2005, preliminary experiments were conducted on breaking seed dormancy in wild *Helianthus* and *Echinacea*. Some promising results were seen for *Echinacea* seed using smoke water and GA3. However, these treatments were not promising for wild *Helianthus* seed.

Computer Application Development:
D. Kovach learned more about using JDeveloper to create client-side and Web-enabled forms, by attending the 2005 Oracle Developer Tools User Group (ODTUG) Conference. He worked with the JDeveloper 10g software and developed a desktop data-entry form for the Amaranth team. This client-side form facilitates entry of seed color, seed shape, and seed starch observation data. The form also allows for barcode scanning of the jar and quick evaluation-method retrieval.

In 2005, an alternative software package for forms development was investigated: Sun's Java Studio Creator. It showed good promise. A web application proof-of-concept was made using this Integrated Development Environment (IDE). The prototype application searched the GRIN database for user-selected characteristic values and also enabled capture of user characteristics and how the requested germplasm samples would be used. Future refinement of this form and where it might be hosted have yet to be determined.

Internet website related:
During 2004, D. Kovach created a new Internet site for the Station and for the Ames Area USDA Civil Rights Advisory Committee's website by using Site Publisher software on the USDA Consolidated Website. The old sites were disabled and new ones established within the mandated completion date of November 1, 2005. A few pages from the old site were reused, new pages created, and all of the Station's Annual Reports were made available on the new site. This website will be enhanced in the coming year.

Cooperative efforts:
In 2005, D. Kovach created or modified Architectural Desktop / AutoCAD drawings to help meet local facility improvement needs. He also supported large-format printing of several professional posters (GEM, Gardner, McCoy, Brenner) and for local needs (image room, germination room, conference room, reception desk room, seed storage room). Other large-format printing support included field planting plans for Pathology and Maize plantings. World and U.S. maps were printed for Seed Storage to display geographic patterns of order distribution. Using JDeveloper and the large-format printer, a large database schema drawing was developed for cooperators at the University of Missouri, who offered to help develop image-analysis software for the Station.

In 2005, M. Erickson worked with J. Carstens and K. Reitsma on two projects. This involved work on *Acer* and *Daucus* seed, as discussed in Germination Testing.

Supervision:
This past year, M. Erickson, Agricultural Biological Science Technician, continued to oversee a crew of four part-time students for help in conducting germination tests.

D. Kovach oversaw the creation of a permanent technician position for germination and the hiring of M. Erickson in that role. They also investigated the requirements for M. Erickson to obtain a Certified Seed Analyst certificate through the Association of Official Seed Analysts (AOSA). They have developed a mentoring relationship with the Iowa State University Seed Science Center to

help prepare M. Erickson for upcoming certification exams.

Germination Testing:
During 2005, M. Erickson and her crew continued to fill seed germination orders, conduct germination tests on over 4,500 accessions, and help curators germinate seeds for their regeneration efforts. This is an increase from last year's level of about 3,600 accessions. In addition to the germination tests that were conducted for the curatorial groups, M. Erickson also carried out two germination tests for the GEM project and a germination test of three *Cucumis* and two *Cucurbita* accessions for a pollinator study. Viability testing statistics are detailed in Table 2.

The germination's crew high level of training and increased proficiency allowed M. Erickson to investigate new methods to promote germination of dormant seeds. M. Erickson dissected several *Daucus* accessions to evaluate variation in the degree of embryo development. She also excised 13 accessions of *Acer* to help determine whether that method would promote germination. M. Erickson performed tetrazolium tests to help determine the seed viability of questionable samples on 30 accessions in 2005. She conducted tetrazolium tests on 24 accessions in 2004; five accessions were tested in 2003.

Germination chambers were managed to accommodate the specific needs of each curator's seed regeneration, as well as to meet our ongoing requirements for periodic viability monitoring.

Plans for 2006:
In accordance with the recommendation of Horticulturist M. Widrlechner and Research Leader C. Gardner, D. Kovach plans a gradual transition from predominately computer application work to seed research work. This may take one or two years, depending on the success of hiring plans for a second IT Specialist.

M. Erickson plans to take the AOSA's certified seed analyst exam for germination this spring, which will be offered at the Iowa State University Seed Science Center.

Seed research priorities will be determined for the coming year in consultation with M. Widrlechner and the curatorial staff.

D. Information Management-Germplasm Collections (R. Stebbins)

Acquisition:
The North Central Regional Plant Introduction Station (NCRPIS) acquired 282 new accessions in 2005. Of these new accessions, 176 were received from within the National Plant Germplasm System (NPGS). This included 78 accessions of wild *Helianthus* from collection trips conducted by NCRPIS personnel and 43 accessions of *Zea* transferred from the National Center for Genetic Resources Preservation (NCGRP).

The remaining 106 accessions, received from outside the NPGS, included 11 accessions of ornamentals and 28 accessions of umbels donated by Hebron University, Israel, and 18 accessions of *Actaea* donated by the University of Massachusetts.

As new accessions are recorded in the Germplasm Resources Information Network (GRIN), an effort is made to include as much passport information as possible. Typical passport information would include a source history, cooperator records, collection-site description, pedigree, secondary identifiers, and any other pertinent information provided by the donor.

Maintenance:
Assistance with curatorial management was provided by processing requests for taxonomic re-identifications and nominations of accessions to the inactive file. In total, 146 accessions received taxonomic re-identifications. Among these were 68 accessions of *Amaranthus* and 38 accessions of *Echinochloa*. Also, 16 accessions were nominated for inactivation, including 7 accessions of *Cucurbita*.

Additionally, 33 accessions were assigned PI numbers. Included in this group were 17 accessions of ornamentals and 16 accessions of *Amaranthus*.

Finally, 2 accessions were inactivated due to duplication. The inventory lots of these accessions were combined with the lots of their respective duplicates.

Projects:
One of the first steps in obtaining a PI number for an accession is to proof the passport information for accuracy and completeness. Proofing passport information is an ongoing project that is secondary to logging in new material. Proofing involves locating paper and electronic files of accession information, corresponding with collectors and donors, and researching Internet sites, maps and GIS databases.

One of my projects involved working with Mark Widrlechner to prepare 17 accessions of ornamentals for PI number assignment. Any errors in GRIN were corrected, and reports were printed for a final check before requesting PI numbers. In addition, I have continued to enter old passport information from logbooks for early Ames-numbered accessions. This long-term project is now 56% complete. During the course of this project, I encountered duplicate accessions and missing GRIN records, which were then corrected.

I assisted Mark Widrlechner to develop and test new accession action codes in GRIN, as part of a system-wide effort to standardize the documentation of passport data and georeferencing.

In August, I staffed an ARS booth in the Agriculture Building of the Iowa State Fair where we interacted with the public and ARS personnel from other units. It was a great opportunity to educate the public about our mission.

Conclusions:
Compared to 2004, new accessions received at NCRPIS were down by 167 in 2005. In maintenance areas, re-identifications were down by 30%, nominations to the inactive file were down by 43%, and PI number assignments were down by 55%, but duplications were double 2004 levels.

All figures for acquisitions and maintenance were below the ten-year average.

E. Order processing (R. Stebbins)

During 2005, there were 1,358 orders entered into GRIN. These orders led to the external distribution of 22,300 items (primarily seed packets, but also vegetative samples) (Table 3A). Of these, 14,956 items (67%) were distributed within the United States, and 7,344 (33%) were sent to foreign requestors. Additionally, 10,428 items (Table 3B) were distributed within the NCRPIS, for such uses as regeneration, evaluation, and germination and pathological testing.

The number of orders entered into GRIN in 2005 was 18% greater than that of 2004; also, the number of items distributed was up by 6,507 or 25%. The number of requests received electronically this year was 1,007, an increase of 26% over 2004.

Cris Nass distributed 481 Initial Accession Performance Report forms in 2005. By September, 235 (49%) had been returned. The Summary Accession Performance Reports and Final Reports that were mailed out in 2005 totaled 60.

F. Seed Storage (L. Burke, L. Pfiffner)

Two full-time, permanent federal employees (Lisa Burke and Lisa Pfiffner), and one part-time, temporary student staffed the seed storage department in 2005.

In 2005, we stored 1985 seed lots, including both newly received lots and those regenerated at Ames or at remote sites. During storage, we bulked 338 lots to create 160 new lots, and 69 original samples were split to create distribution lots due to adequate seed numbers received. Of the lots stored, 1415 were made available for distribution. Inventory records for 3785 lots were reviewed to ensure accuracy of seed amounts, and new labels were printed for lots with outdated labels. We prepared 146 original lots for long-term freezer storage.

Tables 2, 3A, and 3B of the station statistics, found in the appendix, differ slightly from the statistics reported above, in part, due to differences in extraction methods from GRIN. In addition, statistics reported in those tables reflect the collective activities of all Station staff and include both seed and plant inventories.

In 2005, 146 accessions received taxonomic re-identification. All affected seed samples were re-labeled by seed storage personnel. In addition, seed samples of 19 inactivated accessions were removed from the active collection and placed in inactive storage. New labels were made and cold-storage locations and GRIN records adjusted to reflect changes resulting from the assignment of 16 PI numbers.

Seed orders prepared in 2005 included those for distribution, observation, germination, transfer and backup. Seed storage personnel filled 1011 orders. Table 1 of the seed storage report demonstrates the steady increase in seed orders since 1987 (the earliest year GRIN data are complete). Order fulfillment is now tracked with the use of an ORDFILLED action in the order action field of GRIN.
There were 889 lots (840 accessions) sent to the National Center for Genetic Resources Preservation (NCGRP) for backup, including both accessions new to NCGRP and supplemental lots for previously supplied accessions. NCRPIS distributed 25,084 packets to meet distribution and observation requests. Of these, 17,566 were distributed domestically and 7,518 outside the US. We transferred 12 inventory lots to other NPGS sites.

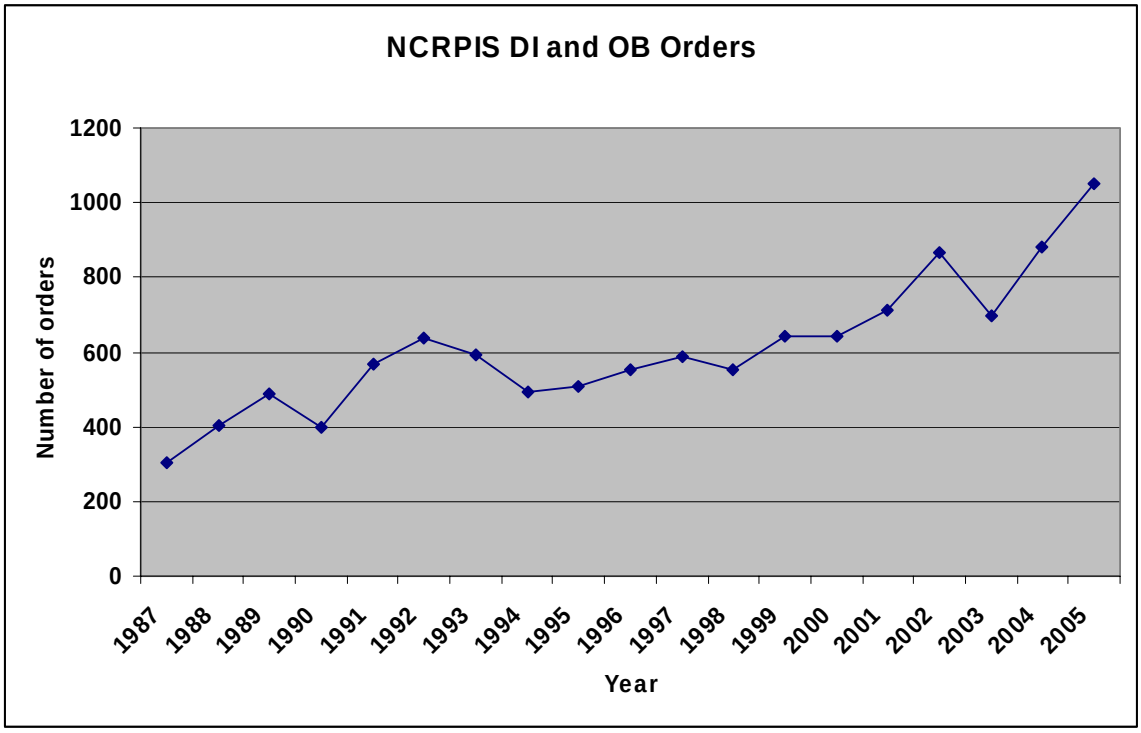

Table 1. DI and OB orders from 1987 thru 2005.

2005 saw the continuation of several projects from previous years. The prepacking program for NCRPIS crops (1,454 lots in 2005 consisting of 20,125 prepacks)increased with the help of our student worker and use of order statistics based on accession use by our customers. Prepacking decreases the amount of time it takes to fill seed orders and helps in maintaining accurate inventory quantities, thus reducing the amount of time required to monitor seed stocks.

In 2005 we also continued training NCRPIS and Ornamental Plant Germplasm Center staff on the new web-based GRIN database and assisting the Database Management Unit (DBMU) with additional testing and improvement of GRIN processes. Lisa Pfiffner, with the assistance of our student worker, completed a large imaging project started in 2004 to document of maize accessions donated by Major Goodman (2,630 packets were inventoried and 5,260 images taken). Additionally, a project was initiated to review the maize reference collection for treated seeds and replacement of old barcode labels (683 lots were marked as treated in the inventory action area of GRIN). Service as resource persons for new curators and the maize project continued with additional assistance from Lisa Pfiffner to the maize project. She served as an official supervisor for student workers on that project due to the absence of a federal employee to serve in that role during much of 2005. Training for 2005 included Blood Borne Pathogen re-certification (Lisa Burke); CPR/First Aid recertification (Lisa Burke) and IT Security Awareness training (Lisa Burke and Lisa Pfiffner).

Plans for 2006:
We will focus on predicting order demand for accessions and developing methods to determine which accessions to prepack in order to increase seed-storage order-filling capabilities.

VII. CURATORIAL AND SCIENTIFIC TEAM REPORTS

A. <u>Controlled Insect Pollination Service Program (S. Hanlin)</u>

Progress:

Caged pollination:

Bee pollinators (minus the experimental alfalfa leafcutter bee) were supplied to 732 cages for controlled pollination of 759 accessions.

Honey bee pollination: Honey bees were used to pollinate 567 accessions in the field and 13 accessions in the greenhouse. Bees pollinated 156 accessions of *Helianthus*., 26 accessions of *Cuphea*, 99 accessions of Cucumis (97 accessions in the field and two accessions in the greenhouse), 46 accessions of *Daucus*, 30 accessions of Cucurbita, one accession of *Pastinacae*, seven accessions of *Ocimum*, two accessions in the greenhouse of *Erysimum*, 78 miscellaneous *Brassica* accessions, 89 miscellaneous ornamentals/medicinal, 36 miscellaneous umbels (35 accessions in the field and one accession of *Celosia* in the greenhouse).

Honey bees were over-wintered in the indoor wintering facility with a survival rate of 95% for the parent colonies and 47% for the nucleus colonies. The survival rate for the nucleus hives was slightly higher than last year's 37%, with the parent colony survival rate being comparable to 2004. This winter, we placed 78 two-story parent colonies, 209 double-story nucleus colonies and 45 single story nucleus colonies into the over-wintering facility. Forty-eight strong three-story parent colonies were left in the field, placing them in groups of four and wrapping each group with tar paper. All queens to be used for queen rearing will be selected in the spring of 2006 from resilient parent colonies.

Queen rearing throughout the summer produced an average of 65 queens per week. Nucleus hives were only produced until late July, because of an increase in unused productive nucs in the field and a decline in the need for late summer honey bee pollination.

Fifty "Italian" three pound packages were purchased and placed into either full size hives or double story nucleus hives. However, the packages had difficulty building up this year and many colonies were placed into nucleus equipment late in the fall for over-wintering. We collected two local swarms which were made into colonies. These colonies were included in our nucleus hive production once they became established and all were over-wintered in the fall.

To prevent swarming in our parent colonies, we removed 45 supers of honey and extracted approximately 450 pounds of honey. The empty supers were placed back on the colonies early in the fall to allow the hives room for late fall stores.

Because of higher mite counts in the fall, all parent colonies and nucleus hives intended for over-wintering were treated. Mite populations were determined using three separate methods of sampling. We used the "powder sugar roll" in which 1 tablespoon of powdered sugar is placed into a jar with 100 bees that are randomly sampled from the hive as our main sampling technique. A second sampling method used this year involved uncapping approximately 25 drone cells per hive, removal of the larvae and examination of the larvae and cell for mites. We also examined for mites on a weekly basis throughout the summer. Ten hives were placed on "screened bottom boards".

All parent colonies and nucleus hives to be over-wintered were fed a total of three feedings of Fumidil-B syrup during the fall. This treatment is for prevention of dysentery in the bees while in the over-wintering room.

For wax moth control during the winter/summer of 2005, stacks of supers containing empty frames were treated with paradichlorobenzene (Para-moth®) crystals on a bimonthly basis to fumigate for moth larvae. Because of the warmer temperatures during the winter, we could not use environmental control methods which consist of opening outside doors on colder days and allowing the room to drop in temperature.

We continued to use our present syrup feeding system of two 1000 gallon polypropylene tanks (one inside the shop and one outside), a 30 gallon "mixing" tank and a dish washer with good success. Minor problems of "crystallization" or crystal formation of the year old syrup in the bottom of the inside tank occurred during the winter of 2005. To prevent this, 100 gallons of water were added to 700 gallons of syrup, and an immersable heater was used for four hours a day and the syrup circulated twice daily.

Bombus pollination: *Bombus* colonies were used in thirteen accessions of ornamentals and medicinal plants (*Cornus*, *Monarda*, *Origanum*, *Diervilla*, *Symphytum* & *Hypericum*); one accession of *Melilotus* was pollinated in the greenhouse.

Six "research" colonies of *Bombus impatiens* were ordered in the spring from a commercial supplier, another two colonies were received in the spring from a cooperator after he was done using them in his greenhouse increases. The bumble bee colonies were used for controlled pollination of thirteen field cages and one greenhouse cage. One Bombus colony can be used for pollinating more than one cage with a minimum lapse of 48 hours between the two locations to prevent contamination. The colonies were checked in the fall to determine if they were strong enough to be over-wintered, however, in all cases they were found to be weak or no longer alive.

Osmia cornifrons/*O. lignaria* pollination: *Osmia* was used to pollinate 82 accessions of *Eruca*, 37 accessions of *Brassica*, five accessions of *Crambe*, three accessions of *Sinapis*, three accessions of *Erucastrum*, two accessions of *Melilotus*, 18 accessions of *Erysimum* (16 in greenhouse and two in field), two accessions of *Iberis*, two accessions of *Matthiola* (in the greenhouse), two accessions of *Euonymus* and one accession of *Isatis*.

Approximately 5,662 bees were used to fill 141 straws in 2005. The total number of domiciles used in 2005 was 278. We collected approximately 1,863 bees last year for use in the spring of 2006. The lower increase numbers resulted due to a cool spring, incorrectly marked purchased straws and early field removal of some domiciles. Because of these low numbers, we will need to purchase bees in 2006.

Fly pollination (McClurg): We continued to use houseflies (*Musca domestica*) and blue bottle flies (*Calliphora* sp.) as pollinators in field and greenhouse regeneration cages containing primarily umbel type plants. They were also used as 'fill in' pollinators in some oilseed accession cages in the greenhouse in the spring of 2005.

We continued to use purchased fly pupae. At the beginning of each pollination season, we determined an appropriate number of pupae to order to meet curatorial needs based on past use and curator-predicted needs. We then arranged for scheduled shipments of blue bottle fly pupae from Forked Tree Ranch, ID every three weeks for winter, 2004-2005 and every two weeks during the summer as these pupae can be held at 0 C for up to three weeks prior to use. We also purchased housefly pupae from J. Coats (ISU, Ames), to use in combination with blue bottle flies for winter, 2004-2005 greenhouse cages. Housefly pupae are picked up from the campus lab on a weekly basis since these pupae can not be stored prior to use.

When both fly species were used in greenhouse cages at the same time, each species was incubated weekly on a staggered basis to ensure continuous presence of flies in greenhouse cages; houseflies were incubated on Wednesdays and delivered Fridays while blue bottle flies were incubated on Fridays and delivered on Mondays. During summer 2005, blue bottle flies were incubated on Tuesday and delivered to cages on Thursday.

Flies were delivered twice weekly to 31 total greenhouse cages from January through 23 May, 2005 for winter/spring pollination season and once weekly to an average of ten cages (high of 23 to low of three) from 25 May through 1 September, 2005 for the summer greenhouse pollination season. Because no umbel accessions failed to bolt in summer 2005 field cages, there were no requests for flies for a fall/winter season in 2005.

An unusual request was made for flies in two spring-blooming field cages of *Euonymous* for Horticulture and eight greenhouse cages of *Erysimum*; flies were supplied from 25 April through 2 May, 2005 but it may have been too cold for them to effect pollinations. Flies were delivered once weekly to a total of 84 summer field cages: 46 cages of *Daucus* (three species), one cage of *Pastinaca*, 35 cages of miscellaneous umbels (13 genera, two with multiple species), and one cage of *Sorbaria* received flies between 27 May through 8 September, 2005. Miscellaneous umbel genera included: *Pimpinella, Zizia, Anethum, Angelica, Coriandrum, Carum, Eryngium, Celosia, Foeniculum, Chaerophyllum, Petroselinum, Ferula,* and *Sium*. One very late blooming field cage of *Matthiola* received flies on 26 September, 2005.

Campus greenhouse fly delivery was performed by horticulture project personnel; entomology personnel delivered flies to farm greenhouses and field cages.

Alfalfa leafcutter bee (ALC) pollination (McClurg):
In addition to research tests conducted for the second year to determine which genera may be successfully increased with the aid of this bee, we used ALC (*Megachile rotundata*) as supplemental pollinators when inadequate supplies of honeybees were available or as a transitional pollinator until a more suitable pollinator or pollination situation was available. The ALC are advantageous to use as pollinators as they are non-stinging and require no care once introduced to germplasm regeneration cages; these bees require about five hours of staff labor weekly for lab manipulations.

Sources for this bee and handling notes are mostly covered under the section of this report pertaining to the ALC research conducted in 2005. The ALC were used in both greenhouse and field cages throughout 2005; genera within the cages that were pollinated by ALC are listed below.

A total of 16 greenhouse cages (nine in the pollinator nest test) containing *Cucumis, Daucus*, *Mahonia*, *Celosia*, and *Helianthus* received ALC bees during winter 2004-2005. Bee introduction started 28 Jan 2005 and continued several times weekly to maintain a bee population of ca 15 bees per cage through 8 April 2005. In mid-February we switched from releasing only bees collected through the emergence box to releasing bees that were maintained with their cells to see if it increased their longevity. As of early April, few bees were emerging from this shipment of cells due to large numbers of parasitic wasps emerging from the cells from that time on; it was necessary to discard the remainder of this shipment of cells mid-May 2005, but fortunately the bees released at the end of March appeared to be remaining alive in the cages by that point in time.

During the springtime, ALC bees were released in 11 germplasm regeneration cages from 29 April through 31 May, 2005 from a second supply of ALC cells. Two early blooming field cages of *Euonymus* received ALC; outside temperatures were probably still too cold for these bees to have effected much pollination. One cage of *Melilotus* and eight cages of *Erysimum* located in the cool greenhouse (heated only to ca 60 F) received ALC bees; these bees were effective in pollinating only

when daytime temperatures in the greenhouse reached 75 F or more with the aid of warm outside temperatures and sunshine.

During the summer, ALC bees were released to a grand total of 213 germplasm regeneration and 15 pollinator test cages from 16 June through 17 October, 2005. Some of the cages received ongoing releases of bees (ALC may not live a long period of time when confined to crops they are not used to, or where only few flowers are available, or due to cool rainy weather), while other cages received only one or two sets of bees until another preferred pollinator could be provided or it appeared the ALC were unwilling or uninterested in the germplasm within a cage and that cage was also receiving other pollinators.

Where the ALC appeared to be effecting pollination in cages or where the curator requested ALC as the only pollinator, releases were made ca every-other-week (as soon as five or fewer bees were noted in a cage) in order to maintain a population of twenty to forty bees per cage (depending on the amount of flowers present in the cage). More bees were released to a cage if: the ALC was the only pollinating insect and there were large numbers of flowers in the cage, the bees were three days old, the weather was cold and rainy.

The following genera were in the three summer greenhouse cages receiving only ALC bees as pollinators: two *Melilotus* and one *Cucumis heptadactylus*. The following genera were in 26 field cages which received multiple releases of ALC as the only pollinator: two *Brassica napus* (in pollinator test), three wild-type *Helianthus* (in pollinator test), six cucurbit and two *Ocimum* and two *Daucus* (in pollinator test), one *Melilotus,* nine *Potentilla* (all different species), and one very-late blooming *Matthiola.*

The following genera were in 189 summer field cages that received ALC once or twice as supplements to other pollinators (usually flies and/or honey bees): 46 cages of *Daucus*, 96 cages of *Cucumis* (47 *C. sativus* and 49 *C. melo*), one *Sorbaria*, two *Alcea*, five *Hypericum*, five *Echinacea*, 21 cages with 17 accessions of nine genera including *Celosia*, *Foeniculum, Coriandrum, Chaeroph, Ferula, Petroselinum, Carum, Melilotus,* and *Angelica* (*Angelica* and *Melilotus* were both noted to be very attractive to the ALC bees), and 13 very-late blooming wild-type *Helianthus* (different species).

In addition, ALC were released to a one-half acre soybean plot for cooperator Reid Palmer (USDA-ARS-CICG, Ames). Ca 1000 bees and accompanying cells were released at three intervals (i.e. a total of at least 3000 bees) from 28 June to 18 July, 2005.

B. Controlled Pollinator Insect Research Program (S. Hanlin, S. McClurg)

Fly pollination of *Pimpinella*:
In cooperation with K. Reitsma and L. Clark, a greenhouse study to determine the following: the species of fly (houseflies only, blue bottle flies only, or two fly species combined), and the frequency of fly introduction (once per week vs. twice per week) was conducted winter through spring 2004. Two species of flies were introduced to 12 of 16 cages of two accessions of *Pimpinella*; seed and data processing were completed February, 2005.

Normally we determine effectiveness of pollination by different insect species by comparing the total amount of seed formed per test cage (or per plant) when germination results for all test cages are comparable. In this test, it appeared that the best comparison for treatments was the percent viability of seed for each cage; this is because *Pimpinella* is considered self-pollinating to some extent. According to the data collected in this study, seed was formed in all cages, but the germination tests showed differences between treatments and between the two accessions. Ames

25735 had much lower viability in all treatments in comparison to Ames 25737 which showed a higher rate of self-pollination. Control cages with no insect pollinators introduced had 0.5 - 21% viability compared to 14 - 93% viability in the insect pollinated cages. Cages of Ames 25735 that received blue bottle flies had 14 - 34 % viability compared to Ames 25737 with 79 - 84%. Cages that received houseflies only or both species of flies together resulted in the highest viability of seed (69 - 93%) for each of the two accessions. The frequency of introduction of flies to cages (once versus twice a week) didn't appear to differ statistically.

Our conclusions from this study:
Self-pollinating *Pimpinella* plants benefit from the addition of insect pollinators. If we are limited to using only one species of fly, it appears the housefly would be a safer choice than the blue bottle fly, but cages of combined species of flies did as well or better than the houseflies alone. It doesn't appear that adding pollinators to the cages twice a week increases the percent viable seed produced in an individual cage.

Alfalfa leafcutters:
We continued to investigate the use of alfalfa leafcutter bees (ALC) (*Megachile rotundata*) as a supplemental pollinator to honey bees with the cooperation of all curatorial staff who utilize insect pollinators. Conditions where the ALC has proved most useful are plants with small to medium size flowers of a flat nature, and warm (26 C or 80 F or above), mostly sunny, dry growing conditions. These bees are preferred by staff that may wish to enter cages frequently to work with the crop since the ALC bee doesn't sting. This bee requires less effort to manage on the part of Entomology staff members as well. To date, the ALC have been used in winter, spring, and summer greenhouse cages of *Daucus, Cucumis, Melilotus*, and summer field cages of the above crops along with *Potentilla*, wild *Helianthus* and miscellaneous umbels. Of the mixed genera umbels ALC were tried on in 2005, they seemed to prefer *Angelica* the most.

We completed some studies begun last year and conducted several different new studies with the ALC bees in 2005. These studies were to determine what we needed to provide these bees to make them more successful in the germplasm regeneration cages. Discussion of our completed studies follows.

Final results of cucurbit/*Brassica* pollination test, summer 2004 field cages:
We received the final seed data from the 2004 field test comparing two treatments (honey bee or HB and ALC) completed in cooperation with K. Reitsma, L. Clark, L. Marek, and B. Bingaman in 2005. Data analysis of five cucurbit accessions (*Cucurbita pepo* and *C. pepo* var. *ozarkana*; *Cucumis sativus, C. melo* subsp *melo*, and *C. melo* subsp. *agrestis*) showed that there were no statistically significant differences between ALC and HB treatments nor the accessions for the following variables: number of fruit, total seed weight, average 100 seed weight. There were significant differences between the ALC and HB for total seed; fewer seed were obtained from the ALC cages. This is due in part to the fact that ALC bees were not available in late July through August when many of the test cage plants were flowering. Data analysis for two species of *Brassica (B. napus* and *B. nigra*) and one *Erysimum* showed there were no significant differences between the quantity of seed formed by HB and ALC for all three accessions.

ALC nest material test in winter 2004-2005 greenhouse cages and summer 2005 field cages:
We wished to determine if ALC would do a better job of pollinating small amounts of non-preferred plant materials (i.e. the diverse germplasm in regeneration cages) and live longer if they were provided nesting materials, both plants for cutting leaf disks and physical structures with the appropriate diameter holes preferred by ALC bees. An additional motivation for adding nesting plant material was so the ALC would not negatively impact the crop increases by removing crop

leaf material for leaf disks, which may cause the crop to put more energy into leaf production rather than seed production. In cooperation with K. Reitsma, L. Clark, L. Marek, and I. Larsen we decided to do an initial test in the greenhouse in the winter of 2004-2005. In addition to crop plants of *Cucumis, Daucus*, and wild sunflower, we provided the following four nesting plants thought to be attractive to ALC for cutting leaf disks for creating their nest cells: alfalfa, buckwheat, commercial hybrid or wild roses, and common mallow which were collected and placed in the greenhouse starting in fall, 2004 so they would be well-established before the test began. Four *Daucus* accessions (*D. carota*) and four *Cucumis* accessions (one *C. sativus* var. *sativus* and three *C. melo* subsp. *melo*) were placed in eight individual cages measuring 3' wide x 9.5' long x 6' high along with one of each of the four nest plant types. Due to lack of greenhouse space for multiple cages, two accessions of wild sunflowers (*Helianthus annuus* and *H. maximilliani*) and one accession of *Daucus* were placed together along with two of each of the four types of nest plants in a single cage measuring 7' long x 6' wide x 5.3' tall. These cages were located in two different greenhouses at the station. In addition, we provided several different physical structures consisting of wood or Styrofoam nest blocks housed in either a wood box or simple wood frame to stabilize the cell blocks in wind or strong drafts in each of the test cages.

The first available bees were introduced to cages on 28 Jan 2005. Because the early-incubated ALC bees do not live for long periods of time and required at least weekly replacement until the end of March, no use of the domiciles or leaf cutting was observed during the initial test period; plant pollination was noted during this time. While the ALC visited flowers on the nesting plant material as well as the crop plants, the nest plant flowers did not appear to distract the ALC from pollinating the crop plants. As of mid-March, ALC were observed moving in and out of nesting blocks in the greenhouse cages, but no leaf cutting was observed until the end of March. It seems that increased daylength (possibly the vernal equinox) was the trigger for this activity. As of mid-April, female bees were actively cutting leaf disks in all cages. The bees cut leaf disks from all plants provided and utilized the *Cucumis* and sunflower petals as well, but roses and alfalfa appeared to be the preferred leaf disk material cut by the majority of the bees. Since alfalfa seemed to be the most practical choice of nesting plant to add to field cages, we decided to utilize that in the subsequent summer field test.

It didn't appear that ALC bees preferred any one domicile design or nest cell material over another, so we chose to use the simplest and least expensive design of wood frame around a solid Styrofoam block (4 inches wide x 5.5 inches long x 3 inches deep with 0.25 inch diameter holes) in the summer field cages.

The summer field plot included three treatments (honey bees, ALC only, and ALC with alfalfa nest plant; domiciles were included in both ALC treatments) applied to six accessions (a single accession each of *Brassica napus, Daucus carota*, and *Ocimum gratissiumum* var *macrophylla*, and three cucurbit accessions *Cucumis sativus* var *sativus, Cucumis melo* subsp *melo* and *Cucurbita pepo*). Initial seed data analysis showed no significant differences between treatments for all crops. We will do a final data analysis in 2006 when the remaining seed data is completed.

Even though the physical domiciles did not appear to make a difference in the pollination effected by the ALC bees, they did provide a place for the bees to spend the night or days of inclement weather; we plan to add domiciles to all cages where ALC are requested as the primary pollinator from here on out for this reason, but will not plan to include extra plant nesting material. It doesn't seem to matter where domiciles are located within cages, but it is best to face them to the south or south-east so they can be warmed quickly by the morning sun to encourage early bee movement.

ALC pollination of *Daucus*:
Normally *Daucus* accessions at NCRPIS are pollinated with a combination of flies and/or honey bees. Studies by researchers in Utah showed that ALC will pollinate onions and carrots. Since it is sometimes difficult to provide honey bees to all cages requested at the same time in summer fields, it seems important to determine if ALC could be used as a supplemental *Daucus* pollinator in our regeneration cages. In cooperation with K. Reitsma and L. Clark we conducted an observational study in several 2004-2005 winter greenhouse cages, using both the nesting test plant cages as well as one of the *Daucus* accessions in a greenhouse cage which is constructed of PVC pipe 21 inches wide x 35 inches long x 44 or 54 inches high and covered with a lightweight fine mesh fabric. We were concerned that the ALC bees may chew through the cage covering, but they did not. The cages were located in two different greenhouses on the station. One of the greenhouses (GH3) is kept at a high temperature (ca 80 to 85 F) and usually has high RH due to the combination of crop plants it houses; the other smaller greenhouse (Entomology) is slightly cooler (ca 75 to 80 F) and lower RH due to the fewer plants it contains. In addition, GH3 may appear darker inside than the Entomology greenhouse due to the differences in construction and orientation of the two buildings. Bees were introduced to greenhouse cages from 28 January through 8 April several times a week as needed in order to maintain a population of ca 15 bees per cage. Bees were observed working the umbels off and on at various times of the day as long as it was a sunny day outside; the bees appeared from 8 AM on in the Entomology greenhouse and from 9 AM on in GH3. Bees were noted working in both greenhouses until at least 3 PM, when observations ceased. The bees were always more active on the *Daucus* in the Entomology greenhouse. There were differences in ALC activity on the different accessions in GH3, the GH3 cages had more flight room above the plants which encouraged the ALC to simply hang on the upper screen near the greenhouse lights, several of the GH3 cages also contained flies and the ALC do not appear to want to interact with flies on the same umbels. In general, our observations lead us to believe that ALC will successfully pollinate *Daucus*, but may not do as complete of a pollination job as the combination of flies and honey bees used traditionally, depending upon the individual accession. One explanation for this is there will be many flies present on a single umbel at one time whereas there is usually only one ALC bee on an umbel at one time. As confirmation of the feasibility of using ALC to pollinate *Daucus*, ALC in the summer field nest test cages were noted to be carrying a lot of *Daucus* pollen (pollen is carried on the abdomen hairs of ALC) where they were the only pollinator.

Our overall conclusion:
We do not recommend replacing the fly/honey bee combination currently used for *Daucus* pollination with ALC, but do recommend using ALC as a supplemental pollinator when flies and honey bees are not available.

GH test of releasing 'bare' ALC vs. bees with cells:
Previously ALC bees had been released into cages with the leaf cells they emerged from when it appeared ca 25% of expected bees were present in the incubation jar; this is also the method that growers use when they release bees to large open plots. We developed the emergence box/collection dish system as concern was expressed about not releasing parasitic wasps which are present in the bee cells to an environment where they may not already be present; in addition, collecting the bees separate from their cells allows us to put a known number of bees in each cage. Some concern was expressed about the amount of staff time it may take to collect bees and the reduced bee longevity that may result with the emergence box system, so we conducted a test in the greenhouse during summer 2005 in order to determine if there was any apparent difference in bee life or activity from the two methods of collection/release. Each of four test cages (two for each of the two treatments) contained the following plants: four pots of buckwheat serving as the flower/nectar source, three pots of alfalfa and two pots of roses (one wild and one hybrid) for leaf cutting. The cages were also provided with identical domiciles. Bees were released on 4 August and 17 August, 2005; 20 'bare' bees and a group of cells with 10 to 20 bees were provided each appropriate cage. Daily

observations were made to determine the number of bees present in each of the cages and to assess their activity. Bee activity from the first release appeared to be more related to the cage position within the greenhouse rather than the bee emergence technique. It did appear that bees released with their original cells may have lived longer in the second release, but numbers of bees present and activity levels were similar for all cages until the test ended mid-September. No conclusive results were obtained from this test and it does not seem important to repeat.

Extended incubation test:
Occasionally some late-blooming plants require pollinators to be supplied to field and greenhouse cages in the late fall to early winter, before a new year's supply of ALC cells would be available for purchase, we wished to determine how late into the calendar year we could incubate ALC cells purchased for summer use; it had been implied that we would get no bee emergence from cells after September. We were successful in getting bee emergence from summer 2004 produced ALC cells through December 2005, although percent emergence was reduced to ca 10%. At of the end of September, some of the bees emerging from cells did appear reduced in size (usually males which are normally smaller than the females), but many of the bees appeared to be of normal size. Some of these later-emerged bees were used successfully in October blooming cages of wild sunflower in the field. The remainder of these test bees were placed with an accession of basil in the greenhouse in the late fall to early winter, but the bees didn't appear to live for more than one or two days; it isn't known if this is because they were not interested in the plant material at hand, or if it was because they were being used outside of the normal time range for their lifecycle and season.

Other observations gleaned from 2005 greenhouse and field cage tests:
Longevity:
We have found during the past two years that ALC bee cells incubated earlier than normal (i.e. January through March) will result in emerged bees, but these bees will have irregular life spans in greenhouse cages until late winter/early spring. It seems necessary to replace ALC bees once to twice weekly in order to maintain a consistent pollinator population in greenhouse cages until the middle to end of March. ALC bees placed in greenhouse cages at the end of March and thereafter seemed to live for extended periods of time (3 - 4 weeks). This was true in both 2004 and 2005 where cages that received bees the end of March still had the same release of bees living and working at the end of April.

We found that in both a field and greenhouse cage of two different *Melilotus* accessions that a 15 day period was required post application before the ALC bees could again work treated plants and remain alive, following use of Ovid/Orthene® pesticide (systemic chemical action) on *Melilotus* for aphid control.

Nest disks/cells:
Because the ALC bee drops slightly (due to the weight of the leaf disk) when they first free a disk they have cut from a plant, the bees seemed to take disks from leaves or petals that were in the most open areas of the cages (i.e. those easily accessible) which makes it easier for them to fly back to nesting block quickly. The ALC were observed carrying leaf disks in *Daucus* field cages where other nest leaf material was not provided; these bees were seen to cut leaves from some weed species, such as amaranth and lambsquarters, that had not been removed from cage interiors.

Although it is not in our plans to collect the cells ALC bees complete in the germplasm regeneration cages for use in future cages (bees from cells produced with the limited food resources inside the cages may be of inferior quality), we did examine cells that were obtained from 2005 cages. The cells created in the winter greenhouse cages were empty; apparently it was too early for the ALC bees to successfully mate and lay eggs. Overall, few cells were filled in the nesting blocks placed into the summer cages with the exception of the *Melilotus* cages; large numbers of cells were also filled in the domiciles placed in the open soybean plot for Palmer. When the nesting blocks from

field cages were hung in an open area to allow bees to continue flying and finish cell development prior to storage in a cooler room in early fall, unknown predators opened many of the cells, leaving few intact cells to examine. The same thing happened to cells from Palmer's open field plot returned to the station on 12 September 2005. If we wished to produce our own ALC cells, we would need to place domiciles in areas with a rich source of food and would also need to bring these domiciles into a protected area as soon as cells appeared to be filled in the block (ca end July to early August). When removed from the domicile, the cells from Palmer's open plot appeared to be highly parasitized; we had been warned that a higher rate of parasitization in cells produced in this area would occur in comparison with those from the western U.S. and Canada. We have not yet tried incubating any of these 2005 increase cells, but cells from Palmer's 2004 open field plot (probably of similar quality to 2005 cells from the same plot) incubated in summer of 2005 did not yield any bees. ALC cells will continue to be purchased from the western U.S. and Canada.

ALC activity around other pollinators:
When ALC are placed in cages where other pollinators (flies and honey bees) are present, the ALC may not pollinate as actively as when they are the only pollinator present in the cage. ALC will compete for pollen and nectar sources that they find highly attractive, but since they are a solitary bee and shy by nature, they may not compete for pollen/nectar sources when other pollinators approach the same flower. In winter greenhouse cages of *Daucus*, ALC seemed unwilling to compete with flies for pollen/nectar. In summer field *Daucus* cages, ALC were observed pollinating the same flowers as honey bees on an accession they found very attractive, however ALC were usually not readily visible in cages that had both honey bees and flies as well.

Release of ALC to open soybean plots:
ALC were observed flying rapidly through the plot, but were never seen working the soybean flowers. It is difficult to observe ALC working flowers with a dense canopy of foliage and observations may not have been made at the proper time of day.

ALC protocols developed or modified in 2005 (McClurg):
Incubation schedule:
Because curatorial staff are not able to predict the flowering of the diverse germplasm accessions they maintain and because ALC bee cells need up to 30 days of warm incubation before all bees emerge from a given set of cells, we set up a continuous bee incubation schedule to ensure an adequate supply of ALC bees will be on hand when needed by curatorial staff. This ALC cell incubation can be started in January or later using cells obtained from the previous summer field season. The first set of bees become available 30 days after incubation is begun. From that point on, a 24 hour notice is needed before these bees can be distributed to cages Monday through Friday.

The actual ALC cell incubation sequence we now use involves three steps for each set of cells set up on a weekly basis:
Step 1. Cells are moved from refrigerated storage to cool room temperature (70 to 75°F) for up to four days of pre-incubation.
Step 2. Cells are moved to a 30°C chamber (84 to 86°F) that is continuous dark for ca two and a half weeks.
Step 3. Cells are transferred to emergence boxes and moved to 30°C chamber that is lighted for five hours daily (8 AM to 1 PM central DST). Cells are left in this chamber on weekdays only and moved to cool room temperature (70 to 75°F) for the weekend. Bees are collected from one set of cells for a two week period.

Purchasing ALC:
The highest quality of ALC cells are obtained from Canadian suppliers. Prior to 2005, we were required to have an import permit to bring Canadian ALC cells into the U.S. The U.S. changed this import policy in late 2004, but we were unaware of this policy change until we experienced

lengthy delays in trying to obtain a new permit; our former permit had expired in late spring, 2004. Once we discovered a permit was no longer necessary, Canadian suppliers were still reluctant to sell us cells without official verification of the new U.S. policy. We obtained a letter from Dr. Wayne Wehling of APHIS, citing the new U.S. regulations which then allowed us to obtain a shipment of cells in early January 2005 for the winter's use.

We purchased two batches of ALC cells from two different suppliers for two different pollination seasons (winter/spring and summer/early fall) in 2005. We have found only one supplier (IPS of Fisher Branch, Manitoba) who will send us cells in December-January for use in the winter greenhouse cages (1 gallon is sufficient for our needs); these cells may have a large number of parasitic wasps (*Pteromalus*) present in them which appear in large numbers in April. We found that a few drops of liquid dish detergent added to the water traps was effective in capturing these pest wasps. We obtained ca 300 bees from 650 cells for the three weeks that cells were placed in the emergence box; the remaining sets of these bees were released to cages along with their cells when there were ca 15 bees present in the incubation jars.

We tried a second supplier (JWM Industries of Nampa, ID with ties to a Canadian site also) for our larger purchase of cells (4 gal received in March) to use for summer cage supplies; these cells appeared to be of superior quality and resulted in emergence of fewer parasitic wasps of a different species. (There are at least 8 total species of parasitoids known to infest ALC cells.) We planned to have bees available for ca 30 cages a week from 1 May thru 30 Sep; from ca 2400 cells incubated weekly, we obtained bees for ca 43 cages (at 20 bees/cage) weekly from 27 April through 17 October, 2005.

Emergence and collection of ALC bees:
Wood bee emergence boxes were modified by adding multiple exits so that more bees could leave the box at the same time; the entrance tubes into the collection dishes were completely lined with plastic tubing which allows the bees to walk unharmed down into the dishes. We began providing Binderboard^{R} wood nest cell layers along with sucrose-soaked cotton wicks to keep the bees calm in the dish until they were collected once daily ca 2 PM. In addition a limited period of light (five hours daily) in the warm incubation chamber also seemed to result in a higher number of healthy bees for daily collection. There is reduced mortality when bees are divided into transport cups the same afternoon they are collected in the dish; this work needs to be done in a cool dark room to reduce the number of escaped bees. Bees are most easily transferred from collection dishes into transport cups, by using the sucrose soaked cotton wicks to scoop up ca 20 bees; if possible, include both male (green eyes) and female (brown eyes) bees in each cup. (Male bees emerge from cells first and females emerge later; there is ca a 2:1 ratio of male: female bees.) Bees that have taken refuge inside the wood Binderboard^{R} cell layers can be gently brushed off into a cup with the tip of featherweight forceps. We found that bees could be held in a cool room (70 to 75°F) rather than refrigerated until taken to cages; emergence boxes are also now placed in the same cool area over the weekend to slow bee activity when no bee collections occur.

Delivery of ALC bees to cages:
ALC bees can be delivered to cages at any time of the day; they will generally be less active before 9 AM and will show greater activity in transport cups on warm days. Bees can be left in transport cups (120 ml or 4 oz. plastic specimen cups with vented lids) at cool room temperature (70 to 75°F) for several days before use, but it is best to use them within 24 hours after collection.

Cool storage:
ALC bee cells are held in a refrigerated chamber to prevent bee development until we are ready to incubate them; cell depth in storage is not to exceed 1.5 inches or bees within the cells will overheat and die. Up to this point, only small quantities of cells were purchased and were divided on arrival into pint glass jars (3 T cells/jar = ca 130 cells) which were covered with pin-prick vented lids for storage and subsequent incubation. With the need to purchase a larger quantity of cells at one time, a more compact method of storing cells was needed. Five 15 inches x 20 inches wood frame trays were made with a copper mesh screen bottom for storage of loose cells; each tray holds ca 5000 ml of cells. There appeared to be no difference in percent emergence of bees held in pint glass jars with vented lids vs cells in the open storage trays in the refrigerated chamber; cells were taken from each of the screen trays and placed into incubation jars at five intervals from 8 April to 15 August and incubated in fall 2005. Bee emergence was checked from 5 to 11 October, 2005 and was similar for all five samples.

Guidelines for assessing ALC bee activity:
We provided the following information to the curatorial staff to aid them in assessing the ALC population in regeneration cages. ALC are warm season bees; they don't show much activity until ambient temperatures hit 80°F or higher. They will be mostly inactive at temps of 75°F or lower; they will not work well in cool rainy weather. These bees may not begin pollination activity until 9 AM or later, so cages should be observed after 9 AM in order to assess the true number of bees present and active in the cage. Cages should be examined for several minutes and from different angles. When ALC are not active on flowers, they will be found everywhere within a cage - on the screen, frame, mulch, and plants. This is true whether a domicile is present in the cage or not. Staff should be careful when they enter cages that contain ALC bees, as it will be easy to step on bees on the ground, knock them off of plants, or squish them between the frame and screen. We place 20 to 40 bees per cage on initial release depending on the following: number of flowers present in the cage, age of bees (add more if bees were collected more than 2 days previously), current weather conditions (more released in cooler rainy weather). We will add additional ALC to cages when we observe 5 or fewer bees flying within a cage. This is a much lower population of bees compared to the several thousand honey bees that curatorial staff are used to seeing, but this small population of ALC will be very effective in pollination. It is important to only release warmed-up new bees to cages, otherwise the ALC already living within the cage may attack and kill the new bees.

Pollination Improvement:
Honey bees:
Several different methods where tested in the spring 2005 in order to fulfill a lack of early nucleus hive demands prior to queen rearing. The first method used was producing several dozen "queenless" hives and allowing the bees to raise their own queen. This method was used in the past to produce early season nucs and to prevent the expenses of purchasing of queens for early nucs, however, it took approximately seven weeks for these nucs to become productive and honey bee pollinator demands were already increasing at that time. The unused nucleus hives were allowed to build up and supers were placed on top so they could be used throughout the summer for nucleus production. The second method tested was to purchase 50 "Buckfast" queens and make two frame nucleus hives with a two frames of brood and adhering bees. This method worked better than the prior technique; however most nucs were not as strong as needed for quality pollination. The final method tested was to purchase three pound packages of honey bees (approximately 3000 bees per package) and 25 "Buckfast" queens. The packages and queens were delivered in late March. The packages were put into double nucleus hives and a week later split into single story nucs with one nuc containing the established "package" queen and the second nucleus hive having a purchased queen added to it. This gave us a stronger nucleus hive than the previous two methods, however in the nucs with newly introduced queens, the population was comprised of more older bees with very few younger bees; this resulted in a slight decline in the bee population before an adequate number of bees were observed pollinating the plants.

A three compartment nucleus hive was built and tested for the mating of queens. Each compartment contained two frames and a separate entrance hole was given to each, however all compartments would share the total bee population. If this method worked, it would allow us to have three mated queens with a minimal amount of bees and frames. The mated queens and frames with adhering bees could be removed and placed into three separate nuc boxes and after a week or so of build up could be placed into a cage. It was found that the number of bees supplied to each nuc only raised two queens successfully; the third queen was either killed or allowed to die in the queen cell.

A commercial "graftless" queen rearing technique was tested twice during the summer. This technique forced the queen to lay eggs in a selected number of cells which were then placed into a queenless hive so the colony would finish taking care of the queen cells. After 12 days, the cells were removed and placed into a queenless nucleus hive where the queen hatched, mated and began her colony. The technique was found to work very well for a minimal amount of queens (20 cells) and would work well if an individual was producing less than 100 queens per year, but was not prolific enough for our situation.

To improve pollination throughout the extra wide duplex cages of *Cucurbita* sp., nucleus hives were placed on wooden boxes so that the bees could fly above the majority of the plant material to get to flowers throughout the cage.

In past years, we have had problems at one of our permanent bee yards in the spring and fall with raccoons pulling apart the boxes and eating the bees and honey from the frames of nucleus hives. A 4-strand/two controller electric fence was placed around the hives at this location in the spring after we started seeing damage occurring. The fence worked well over time in deterring the raccoons; in the future, it will be placed out in this yard prior to hive introduction.

Osmia:
A yearly problem in placing "increase" *Osmia* domiciles in the spring is difficulty finding them later in the summer after the trees have filled out in vegetation. This year as a possible solution to this problem, a handheld GPS unit was used in the spring when placing the domiciles; the coordinates for each domicile were recorded. In the summer when it was time to retrieve the domiciles, the hand held was used again to accurately find them. All domiciles were located this year and in less time than in previous years. Because of the success of this method, the use of this device will be a regular aid when putting out and collecting the annual "increase" domiciles.

To possibly eliminate discarding still viable *Osmia* bees once a domicile is removed from a field cage for spraying, we tested a method in which domiciles were relocated to the fringes of the field before chemical application and then placed back into the same cages at the time specified for re-entry. The domiciles were removed from and placed back into cages only during the early morning hours. Some bees were observed flying into and out of the domiciles while they were located at the field edge, but mowing of these areas contributed to loss of forage for the bees. Some domiciles were also lost to field vehicle traffic when their presence wasn't noted. The results of this trial were varied; in some cages bees were observed continuing to pollinate the plants within following reintroduction, while in other cages the domiciles had to be replaced with a new domicile containing straws from which bees had not yet emerged. We found that we should not have used the same location within the cage when re-introducing the domiciles, as many of the domiciles were knocked down in storms which followed. Next year this reintroduction method will be retested, placing the domiciles at several locations along established fence lines which should have fewer disturbances from human activity.

Pollinator Protocols:

Request form:

An electronic "pollinator request" form was used throughout the summer of 2005. This form made pollinator requests more uniform from curatorial staff and pollinator distribution more equitable. This form also allowed the entomology staff to obtain more accurate records of the cage numbers needing pollinators, the date when pollinators where requested, the total number of cages needing pollinators, and better organize their activities.

Pollinator tips:

We began presenting pollinator tips at staff meetings to clarify to curatorial staff how to request pollinators, understand insect behavior, and perform plant-related tasks such as watering and chemical applications in order to result in the least harm to the insect pollinators present in the regeneration cages in the field and greenhouse. Topics covered this year included: fly schedules and protocols, how to water greenhouse cages, generalizations about insect development and life cycles, Osmia - the spring bee, how often and when to request each of the available pollinator species, demonstration of the electronic pollinator request form, how to determine if additional ALC bees are really needed in a cage, bee stings and anaphylactic shock, making honeybee nucs.

Operations manual:

The "pollinator section" of the NCRPIS operations manual was reviewed and updated to show the revised or improved methods of rearing or emergence of insects used as pollinators. The section was also updated with explanations of additional insects now used as pollinators such as fly species and the ALC bees.

NCRPIS internet website:

The "pollinator links" for the internet website were updated with recent pictures taken by the entomology staff, revised information of how the pollinator are used at NCRPIS and updated methods for rearing and emergence of the insects being presently used.

Cooperation (S. Hanlin):

A cooperative study was carried out in July and August with Dr. Reid Palmer, USDA-ARS, and his graduate student Evelyn Ortiz-Perez. The objective of the study was to continue to observe the attractiveness of soybean flowers to alfalfa leafcutter bees and honey bees and to try and determine the nectar makeup in the accessions most attractive to pollinators. A total of five ALC domiciles were placed on the north side of a one-half acre plot with entrances facing south. Four total double story nucleus hives were placed on both the north side and in the center of a second one-half acre plot with the entrances also facing south. Dr. Palmer and his assistants made all plot data observations and nectar makeup analysis. The NCRPIS pollination staff supplied all insects needed in the project and the expertise for determining needed domicile numbers.

The entomology staff assisted Mark Smith, ISU forage breeding project, with recommendations, placement and emergence of greenhouse pollinators. Mr. Smith was shown several of the cage designs used to hold greenhouse pollinators used at the station and then designed a cage to use. Mr. Smith was given information on Bombus suppliers and assisted with placement of the hives in his cages. A single story nucleus hive was supplied to Mr. Smith for use in pollination of alfalfa in a greenhouse cage. More recently, alfalfa leaf-cutter bees were emerged and supplied to Mr. Smith for use in pollination of birdsfoot trefoil over several weeks. Mr. Smith quit using ALC because of not being able to keep his greenhouse at a preferred temperature for the bees and started using Bombus once again.

A cooperative exchange of information on the emergence of ALC occurred with David Bell of NTFLGCR in Prosser, WA. Mr. Bell has designed a piece of equipment in which the pupae is introduced, the parasitic wasps are collected/destroyed and the adult bees are collected and placed

into cages. The total number of bees used at Prosser compared to Ames was also discussed in addition to what crops both locations are using the bees to pollinate.

Heathcliffe Riddell, a postdoctoral associate at the University of Wisconsin, requested protocols and recommendations on the use of ALC for caged clover pollinations. In the summer of 2003, Dr. Riddell had used several cages at the station and the assistance of the pollination staff with his graduate work with clovers at ISU. Dr. Riddell was sent the emergence protocol which is presently being implemented at the station and a list of possible vendors of ALC bees and supplies.

Entomology activities and protocols were discussed by both McClurg and Hanlin with A. Gulbani, visiting scientist from Republic of Georgia in March 2005.

Presentations:
On April 2, S. Hanlin spoke at Reiman Garden's "Kidz-day". The presentation was on honey bees and their use as pollinators of plants with a focus of how they are used at the PI station. Approximately 75 grade school age children and their parents listened to the talk.

On May 12, S. Hanlin was invited to present to approximately 200 sixth graders at the Squirrel Hollow Nature Center "outdoor classroom" in Jefferson IA. The hands on presentation was based on general honey bee facts, the equipment used in beekeeping and a short introduction to the other pollinators used at NCRPIS. On the day of the field day, the weather forced a cancellation of this program; however an invitation was extended to participate in 2006.

On July 16, S. Hanlin organized and participated at the Iowa Honey Producers "field day" at NCRPIS. Approximately 50+ individuals participated. The focus of the meeting was a presentation by Gerry Reynolds, a Kansas beekeeper, and his experience with the Russian bees and their resistance to parasitic mites. Mr. Reynolds's research could possible be a precursor to a research project which has been discussed in cooperation with the USDA Baton Rouge Bee Lab.

On July 25, S. Hanlin, N. Behrens, and the Iowa State Honey queen spoke at the Iowa Family and Consumer Conference in Ames. The focus of the presentation was on honey bees and their products based on a nutritional value. Several participants were interested in the use of pollinators at NCRPIS and there were several inquiries about several of the genera grown at NCRPIS and their current use in the U.S. market such as Amaranthus and Echinacea.

On December 7, the entomology staff presented a power point presentation at the NPGS Curator Workshop in Chicago, IL. The presentation was titled "The Art of Pollination in Cages with Insects". We discussed each of the six insect pollinators used at NCRPIS, including: costs, specific crops pollinated, and the techniques used for rearing and introduction of the pollinators into the cages.

Research Plans for 2006:
In cooperation with L. Marek and B. Bingaman, we plan to conduct a comparative study using ALC, flies (house flies and/or blue bottle flies), and honey bees or *Osmia* (depending on how late it is in the spring). This will be a caged field study using three genera of *Eruca sativa*. The purpose of this study is to determine if ALC pollination of *Eruca* is comparable to the other pollinators which have been used in the past. It is also beneficial to know if ALC can be used to replace the diapausing *Osmia* in the summer rather than honey bees which have been used in the past. Flies will be included to assess the apparent success of the seed corn maggot fly which was an unintended pollinator present in some of the 2005 oilseeds field cages.

In the *Osmia* removal/replacement study done in the spring of 2005, we only observed adult bees in approximately half of the cages which had domiciles reintroduced into them. A possible problem

was that the field fringe where we placed the domiciles in the interim during spray application was mowed on one occasion and several domiciles were knocked over by field vehicle traffic. Also it was observed that bees appeared to be disoriented because of few landmarks and thus were lost. When we remove the domiciles in summer 2006, we will relocate them to one of two locations, either near the fenceline near the woods on the north side of Mortenson Rd. or just north of highway 30 along the fence on the southern part of the station.

We plan to repeat the extended incubation/emergence of ALC bee cells in 2006; we will provide plants of known interest to the ALC bee (*Cucumis, Daucus*, wild-type sunflower) in greenhouse cage to determine if the bee will remain living for longer periods in the fall/winter.

To give us more accurate records when placing pollinators into cages, S. McClurg plans to test a handheld bar code reader for collecting the date, cage number, accession number and the genus/species for the crops. S. Hanlin will cooperate with her to test if this system would work for collecting and holding information from the individual bee yards in addition to placing pollinators in cages. This system should give more accurate records to pass on to the curatorial staff in a more immediate manner.

In order to improve pollinator effectiveness in the large *Cucurbita* cages, we are going to continue to raise the honey bee nucleus hives above the canopy of foliage by placing them on supers or boxes. We are also going to test the use of two honey bee nucleus hives per cage, one at the north end and one at the south end of each of the cages. In addition we will try using Bombus as a pollinator in one of these large cages to determine if they are a more effective pollinator than the honey bee in this situation.

Publications/Posters:
Evaluation of Insect-mediated Seed Set Among Soybean Lines. E. Ortiz-Perez; H. T. Horner; S. J. Hanlin; & R. G. Palmer. Field Crop Research.

C. Plant Host-Insect Resistance and Control of Seed Infestation Research (S. McClurg)

Progress:
Field:

Sunflower resistance to sunflower moth: It was determined in spring 2005 that there were inadequate numbers of intact samples from the accessions grown in the field summer, 2004 for accurate data assessment. These samples were damaged by crows immediately prior to field harvest and a large quantity of mice allowed into the dryer bin by a malfunctioning controller in October, 2004.

Final data analysis of all core accessions tested from 2001 - 2003 needs to be completed and provided to sunflower curator Laura Marek for entry into GRIN.

Sunflower moth: The laboratory colony of sunflower moths reared at NCRPIS for over 20 years was discontinued in April, 2005 since no further field evaluations were planned.

Laboratory seed investigations:

Coriander: I continued to work cooperatively with curator D. Brenner and Agronomist D. Kovach to assess the percent chalcid infestation in coriander increased in Ames field plots; I dissected samples of eighteen accessions in February 2005 which had been grown in summer 2004 field plots. In March I dissected an additional two samples that had been

treated with liquid nitrogen, to verify that treatment successfully kills the chalcids infesting the coriander. It was determined that chalcid larvae killed by liquid nitrogen treatment simply turn brown but maintain their large size until exposed to air after which they will begin to disintegrate; they can be visually distinguished from larvae that have died of other causes which are usually shriveled when found in seeds.

A non-destructive method of assessing coriander germplasm increases for the presence of chalcid larvae with accuracy similar to that of physical dissections was needed. It was suggested that we examine digital images of X-rayed seed; I determined that digital images of coriander which were computerized scans of radiographs produced from the NCRPIS Faxitron X-ray Cabinet system, model no. 43855A, purchased ca 1985 could not be successfully read for percent infestation by chalcid larvae. The recently purchased Faxitron MX-20 X-ray system at the OPGC, OH produced quality digital images that showed insect infestation within coriander fruits. Fifteen total seed samples (two replications of three accessions with two treatments (an untreated control and liquid nitrogen for ca 16 hours) along with one 'practice' set for each accession) were sent to OPGC personnel in trays so that individual seed image results could be compared and verified with physical dissections completed in Ames.

Some general comments about discerning the contents of the fruit in the digital X-ray images: If the coriander fruit was either on it's end or "offset" when x-rayed, it was easier to tell what was in both halves of the fruit. If the fruit was on its side when x-rayed and the top half contained a good embryo, it was not possible to determine the contents of the bottom half of the fruit. If an empty seed was on the top side in such a situation, it was feasible to determine the contents in the bottom half of the fruit. It was found that wasp exit holes do not show up in X-ray images. It was also discovered that it was not entirely clear in the digital image what life stage of the insect was present in an infested seed, or if in fact there was still an insect in the seed. I also found that it is easy to mistake an underdeveloped seed for an infested seed in reading the digital X-ray images.

Detailed image analysis (the orientation of the seed or fruit in the x-ray and the apparent contents of seed: good embryo, empty or underdeveloped embryo, infested, or unknown) was done 12 – 13 April 2005 and percent infested seed was calculated. In addition a quick count of infested seed per image was also recorded and compared to the detailed analysis; the quick count included an initial count of the number of individual seed visible in the entire image and a count of the number of seed that appeared to be infested. In the two accessions with low percent infestation this quick visual count appeared to be as or more accurate in assessing the actual percent infestation as determined by dissection than that achieved with more detailed analysis of the digital X-ray image. The quick assessment was less accurate in determining percent infestation in the highly-infested accession, however.

In order to compare the accuracy of percent infestation calculated from the image to the results of percent infestation determined by dissection, it was recorded for each seed whether the results of the dissection were the same as or different from the image determination. Where "infested" was recorded for the image seed contents, and there was either an exit hole or a form of the chalcid present in the seed when dissected, a "qualifed yes" was recorded for accuracy. Since the orientation of seed/fruit in the X-ray obscured the true contents of some seed in some of the fruits, the number of matches (yes, qualified yes, no) were recorded separately for the two seeds in each fruit. The exact (yes only) and qualified percent matches or accuracy were calculated; it was found that the use of the "qualified yes" only made a difference in the highly infested accession. In all samples, the percent accuracy (exact or qualified) of reading X-ray images was 75 – 91% compared to the

results of dissection. It appears this is an acceptable enough level of accuracy for the curator.

Brenner requested that this coriander pest be identified to species if possible. Two parasitic wasp experts were contacted, who subsequently identified our pest as *Systole coriandri*.

I assisted in writing and reviewing the appropriate materials and methods section of the paper *Liquid Nitrogen Controls Seed-borne Chalcids Without Reducing Germination in Coriander Seeds* in cooperation with Kovach, Brenner, Gardner, and Widrlechner. This paper has undergone peer review and has been submitted for publication consideration by Kovach.

Contacts:
S. McClurg was asked in March 2005 to respond to a 'Rearing specialist survey'; data provided was used for a presentation at Eastern branch ESA meeting given by Dr. Norman Leppla, U of FL.

In May 2005, S. McClurg provided information to M. Lopez of the USDA-ARS-CICG, Ames, regarding work former research entomologist R.L. Wilson did with amaranth diets for European corn borer.

Plans for 2006:
I will continue to support activities for the NCRPIS insect pollinator program, assisting in the incubation and placement of bees and flies, as well as providing cooperation in pollinator studies proposed by S. Hanlin and NCRPIS curators. Continued investigation of the alfalfa leafcutter bee is a priority.

I will work with the sunflower curatorial team to determine extent of sunflower seed weevil infestation in perennial wild-type sunflower plots and assist in the investigation of control alternatives; continue to offer support to all NCRPIS and GEM project personnel when insect pest identification/information is needed; and continue to process, archive and make data publicly available from past host plant resistance evaluations.

I will work with other NCRPIS staff in development and implementation of barcodes on field and greenhouse cage labels to make pollinator delivery data entry more accurate and efficient.

I will also work on indexing past Entomology project slides and photos to make them accessible to other NCRPIS staff.

Summary of Pollinators supplied to Greenhouse and Field Cages, NCRPIS, 2005
Specified are number of cages and plant genus for each pollinator and curatorial project.

Season	Location	Project	Honeybee	Osmia Bee	Bumblebee	ALC bee	FLIES
Winter-Spring	Greenhouse	Brassica	2 Erysimum	16 Erysimum		8 Erysimum	8 Erysimum
		Horticulture	8 miscellaneous		3 Cornus	1 Mahonia	3 Hyoscyamus 1 Malva
		Misc Umbel	1 Celosia	2 Melilotus	1 Melilotus	1 Celosia 1 Melilotus	
		Sunflower				2 Helianthus(2poll)	
		Vegetable	2 Cucumis			7 Cucumis (4 poll) 5 Daucus (4 poll)	27 Daucus
			13	18	4	25	39
Spring	Field	Brassica		82 Eruca 37 Brassica 16 misc.			
		Horticulture		2 Euonymus		2 Euonymus	2 Euonymus
				137		2	2
Summer	Greenhouse	Misc Umbel				2 Melilotus	
		Vegetable				1 Cucumis	23 Daucus
						3	23
Summer	Field	Brassica	74 miscellaneous (1 poll)			1 Matthiola 2 B. napus (2 poll)	1 Matthiola
		Horticulture	33 miscellaneous		4 Diervilla 2 Monarda 3 misc.	9 Potentilla 2 Alcea 1 Sorbaria	1 Sorbaria
		Medicinal	56 miscellaneous		1 Hypericum	5 Hypericum 5 Echinacea	
		Misc Umbel	35 miscellaneous			3 Melilotus 19 miscellaneous	35 miscellaneous
		Sunflower	156 Helianthus 26 Cuphea			16 wild Helianthus (3 poll)	
		Vegetable	127 cucurbit(3poll) 46 Daucus(1poll) 8 misc. (1 poll)			102 cucurbit(6poll) 48 Daucus (2 poll) 2 ocimum (2 poll)	46 Daucus 1 Pastinaca
		R. Palmer*	4 double story nucs open soybean plot			3 large releases open soybean plot	
		V. M. Cruz	2 nucs - brassica				
			567	155	10	215	84

* R. Palmer – USDA-ARS cooperator

D. Plant Pathology (C. Block, B. Van Roekel)

Research Notes:

Maize (*Zea mays)*:

Three hundred and ninety (390) maize accessions, mainly inbred lines, were evaluated for Stewart's wilt (*Pantoea stewartii*) resistance. This was an excellent trial with good differentiation among accessions. The five most-resistant accessions, with average scores of 2.0 or lower on a 1-9 scale, were Ames 23426 (A357), PI 550549 (IA High Oil Synthetic), PI 558533 (Mo21R), PI 601008 (PHG35) and PI 601229 (PHG83).

A real-time PCR assay was developed, in conjunction with Anania Fessehaie and Lisa Shepherd of the Iowa State Seed Science Center, for detection of *P. stewartii* from maize seed. PCR primers and TaqMan probe were designed from DNA sequences in the 16S-23S rDNA intergenic spacer region. Research is underway to evaluate the sensitivity and reproducibility of the assay for detecting the pathogen from infected seeds.

Sunflower:

Sixty-nine wild sunflowers accessions were evaluated in a second year field trial for Septoria leaf blight resistance (*Septoria helianthi*) to correlate geographic origin and disease resistance. Resistance to Septoria leaf blight was clustered in a group of accessions from the Gulf Coast of Texas and also found in accessions from the Midwest. The best performing accessions from Texas were PIs 435419, 435421, 435427, 435428, 468511, 468514, and 468517; from Kansas, PIs 586856 through PI 586864; from Illinois, PIs 435615, 478307, 547167, and 547168, and from South Dakota, PI 597893.

Amaranthus:

We cooperated in a greenhouse trial with D. Brenner to evaluate *A. tricolo*r selections for *Phomopsis amaranthicola* resistance. No high levels of resistance were found, but some breeding lines show improved resistance. See amaranth curation report for more detail.

Cucurbits:

We continue to evaluate protocols to prevent bacterial fruit blotch (BFB) spread in GH cucurbits. See vegetable curation report for more detail.

A small GH study of within-plant movement of squash mosaic virus (SqMV) showed that the virus moved from an inoculated leaf near center of plant to the growing tips (leaves and fruits) within 7 to 10 days. Movement from one leaf to adjacent leaves was much slower than movement to the tips. The rapid within-plant spread highlights the importance of using screened cages in the field to exclude cucumber beetles, the main vectors.

Disease observations on seed increase crops:

Field observations for plant diseases were made in the seed increase plots of *Brassica*, cucurbits, sunflower, carrot, basil and maize.

Brassica and related *Brassicaceae* genera:

Seed regenerations plots in field W2 and N11 (194 accessions) were surveyed for diseases in mid-June. Diseases of interest included black rot (*Xanthomonas campestris* pv. *campestris*), blackleg (*Leptosphaeria maculans*), powdery mildew (*Erysiphe cruciferarum*), Alternaria diseases (*Alternaria* spp.), downy mildew (*Peronospora parasitica*) and white rust (*Albugo*). Black leg, downy mildew, and *Alternaria* diseases were not found in 2005. White rust was moderately heavy on one accession of *Lepidium latifolium* (Ames 21390). Powdery mildew was found on one *Lepidium latifolium* (Ames 15720), one *Crambe filiformis* (PI 388866) and one *Crambe kralikii* (PI 388797).

Black rot (Xcc) was found in 14 of the 194 cages. The disease was present at trace levels except in six accessions where 25% or more of the plants were infected.

Cucumber and melon (*Cucumis sativus* and *C. melo*):
Six disease surveys were conducted in field N9 between 20-June and 01-Sep-2005. Bacterial fruit blotch (BFB) was absent during 2005 in all 94 accessions. Angular leaf spot, caused by *Pseudomonas syringae* pv. *lachrymans* was prevalent on June 20 and caused considerable leaf damage on a few accessions. The disease faded away as temperatures warmed up and was barely detectable by mid-August. Anthracnose (*Colletotrichum orbiculare*) and powdery mildew showed up late in the season.

Sunflower (*Helianthus annuus*):
The main disease of phytosanitary importance is downy mildew, caused by *Plasmopara halstedii*. Sunflower field inspections were conducted for downy mildew, viruses, and aster yellows. In 2005, one downy-mildew infected leaf was found and the plant was removed.

Corn (*Zea mays*):
The maize seed increase plots (155 accessions) were surveyed during late August for the presence of Stewart's wilt, common rust, common smut, gray leaf spot, and northern leaf blight. Stewart's wilt did not show up until about mid-August. Northern leaf blight (*Exserohilum turcicum*) was more abundant than usually found. Lab tests were done on 490 corn seed lots for Stewart's wilt, 10 accessions for *Stenocarpella maydis* and three accessions for *Helminthosporium* diseases.

We identified Macrophomina charcoal rot on corn ears from increases on the island of St. Croix. Although the pathogen has been reported as present on soybeans in Iowa, infected seeds and cobs are being collected and disposed of by autoclaving.

Carrot and basil:
The carrot cages were surveyed on 5-July-2005. The only widespread problem was an unusually heavy level of powdery mildew in 23 of the 46 cages. Flowering was nearly finished and seed yields were high, so no control action was taken. The seven basil accessions had no diseases of note.

Cucurbit virus-testing:
Greenhouse-grown cucurbit seedlings were tested by ELISA for seed-borne squash mosaic virus before transplanting. Three of the four infected *C. pepo* accessions were original seed samples (new to the station). Infected plants were removed from the population before transplanting. Test results are summarized in the following table.

Species	Accessions tested	Accessions with infected plants	Plants tested	# of SqMV infected plants	% infected plants
C. sativus	50	0	1138	0	0%
C. melo	47	1	1586	4	0.2%
C. pepo	32	4	1189	26	2.1%
Total	129	5	3913	30	0.7%

Miscellaneous:
We isolated and identified the first occurrence of anthracnose, caused by *Colletotrichum gloeosporioides*, on St. John's Wort (*Hypericum*) plants at Plant Introduction farm. This is a devastating stem disease of *Hypericum* that causes rapid plant death.

Publications:
Block, C.C. and Reitsma, K.R. 2005. Powdery mildew resistance in the U.S. National Plant Germplasm System cucumber collection. HortScience. 40(2):416-420.

Block, C.C. 2005. Evaluation of wild *Helianthus annuus* for resistance to Septoria leaf blight. Proc. 27th Sunflower Research Workshop, Fargo, ND, Jan 12-13. Online. http://www.sunflowernsa.com/research/research-workshop/documents/Block_Septoria_05.pdf.

Menelas, B, Block, C.C., Esker, P.D. and Nutter, F.W., Jr. 200X. Quantifying the feeding periods required by corn flea beetles to acquire and transmit *Pantoea stewartii*. Plant Disease (in press).

Plans for 2006:
1. Continue long-term evaluations in the maize collection for disease resistance - Stewart's wilt and/or northern leaf blight.

2. Conduct research on the detection sensitivity of the real-time PCR assay for *Pantoea stewartii* from maize seeds. Work is also underway on a real-time PCR assay for *Stenocarpella maydis* (Diplodia blight of maize) to be combined in a multiplex format with *P. stewartii*.

3. Conduct greenhouse/growth chamber experiments, in collaboration with Tom Gulya (USDA-ARS, Fargo) to improve a greenhouse protocol for assessing sunflower disease resistance to *Sclerotinia sclerotiorum*. Pending grant funding, this project would expand to screening wild sunflower germplasm.

4. Review the blotter method seed health assay for detection of *Leptosphaeria maculans*, (anamorph *Phoma lingam*) from *Brassica* seeds to evaluate the conditions needed for optimal detection/recovery of the pathogen.

5. Assess long term survival of the bacterial fruit blotch (BFB) pathogen in melon seeds via GH grow-outs of old seed lots.

E. *Amaranthus, Celosia, Chenopodium, Coronilla, Dalea, Echinochloa, Galega, Marina, Melilotus, Panicum, Perilla, Setaria, Spinacia* and miscellaneous Umbelliferae and Poaceae (David Brenner and Sam Flomo)

Acquisition and inactivation (Table 1):
A new amaranth grain cultivar Raudonukai was received from Lithuania. It is black seeded, which is unusual for grain types, and had high yields in Lithuanian trials.

Fifteen new umbels were received of landrace types of anise, coriander, and cumin from the Palestinian Territory.

Maintenance and distribution (Tables 2, 3A, and 3B):
In Table 2 the differences between the #Attempted and the #Harvested columns are chiefly due to the fact that the #Attempted includes plantings for observation, when harvest was never intended. In 2005, 235 plantings were for observation (mostly taxonomic) and 188 were for regeneration. Osmocote™ slow release fertilizer was used to fertilize the soil for our caged seed increases. Previously, fertilizer was applied to whole fields using large farm implements. Hand-fertilizing allows us to adjust rates in small areas; it is desirable to limit some plants' growth by fertilizing them less. This also seems to reduce the weeding needed since difficult to weed areas at the cage edges are unfertilized.

In general, it was a good field season and harvests were adequate.

Amaranthus and *Chenopodium*:
The 212 accessions ordered for observation were primarily for taxonomic determinations in greenhouse plantings at the NCRPIS. The regenerations were grown in the fall greenhouse.

Echinochloa, *Panicum*, *Setaria*, miscellaneous Poaceae:
Ten *Tridens flavus* accession performed well in field cages.

An *Apera intermedia* accession performed well in the winter greenhouse after an earlier failure in the field. *Apera* are winter annuals and are better suited to our winter greenhouse than to our field conditions.

Melilotus and other legumes:
Two winter annual accessions from the Mediterranean area performed well in the winter in GH-2 after failing in the field. Based on this experience, it is thought that other minor species of Mediterranean annuals may prosper in the greenhouse. This seasonal adaptation is recorded in the Sweet Clover GRIN descriptors under duration.

A *Melilotus* accession (PI 629290) was found to have an unusual growth cycle. It had strong vertical growth in the first year, a common characteristic of the bolting annual type, but required vernalization to flower as a typical biennial. It is a laboratory research line that was provided by Dr. Herman Gorz, then of the USDA-ARS in Lincoln, NE. The trait is documented in GRIN in the duration descriptor.

Spinacia:
An accession of wild *Spinacia tetrandra* from Georgia (Ames 26359) germinated better than in our previous experiments. The seeds were planted in January 2005 in sand. Sixty one seeds germinated from September to November 2005. In a plastic greenhouse that is cool in the winter and warm in the summer, with substantial temperature swings depending on the weather. This method is based on a similar method published for *Chaerophyllum*:

> Baskin, C.C., T.S. Hawkins, and J.M. Baskin. 2004. Ecological life cycle of *Chaerophyllum procumbens* variety *shortii* (Apiaceae), a winter annual of the North American Eastern Deciduous Forest. Journal of the Torrey Botanical Society 131:126-139.

It seems that wild *Spinacia* seeds, like those of some other winter annuals, require a long, warm-humid dormancy breaking treatment resulting in synchronized germination in the fall. This would represent substantial progress in dormancy-breaking methods development since seed dormancy is the largest obstacle to maintenance of wild spinach germplasm. If successful, the method will be published in an appropriate journal so that it can be investigated for potential application to other seed dormancy challenges.

Seed regeneration by collaborators in Sakata Seed Inc. and USDA-ARS in Salinas California resulted in 13 harvests in early 2005. A new planting was not started for 2006 harvest because very few accessions need regeneration.

Miscellaneous Umbelliferae:
The field plantings of *Coriandrum* had very low frequencies of Chalcid (*Systole*) seed infesting insects this year. This improvement could be due to the presence of a smaller planting that attracted and nurtured a smaller population of chalcids than in other years, but this is speculation. The severity of chalcid infestation has been inconsistent. The recent low frequency of infestation is a very welcome improvement over the years of heavy infestation.

Field plantings of *Foeniculum* and *Sium* and were successful. Most of the *Angelica* plantings did well, including some that over-wintered in the field. One *Angelica gigas* (Ames 24084) with dark purple flowers was destroyed by spider mites during flowering.

Viability testing:
It was a terrific year for viability testing thanks to Maria Erickson and her group. Fourteen percent, or 1,227 of our 8,664 accessions were tested (Table 2).

Of those tested, 567 were *Amaranthus* accessions. Twenty-two *Amaranthus* accessions became un-available because of low germination as demonstrated by this new data. Most of these cases (18 accessions) had low germination in 1988 tests and higher germination in 1991 tests. Most of the low germination seed lots were field-grown in 1987 inside of paper pollination bags, which may contribute to mold infestation of seeds. Greenhouse methods presently in use have yielded healthy seeds.

Characterization/taxonomy/evaluation (Table 4):
I traveled to the University of Illinois to copy passport data records from the Crop Evolution Laboratory, courtesy of Ted Hymowitz. This new data is safely filed but not yet entered in GRIN. Five-hundred-thirty-four accessions held at the NCRPIS have "UI" numbers and information associated with their records can be improved from this effort. Most of these were donated to the NCRIPS from the Crop Evolution Lab in the 1980s.

Five new *Echinochloa* descriptors were written and deployed in GRIN. Two hundred fifty-three observations were loaded into these new descriptors.
I made 111 taxonomic changes in 2005 for my combined crops. The majority of these (67) were in *Amaranthus*. We are near the end of a multi-year effort to resolve the taxonomy of the *Amaranthus* sp. accessions in the collection. In *Echinochloa,* 37 were changed, most of these were very long standing issues. The remaining six changes were in the Umbels, grasses, and *Melilotus*.

Enhancement and/or utilization:
Amaranthus:
Two ornamental varieties from my breeding efforts were released by Iowa State University: 'Pillar Orange' and 'Pillar Red'. By comparison to older ornamental types they have lodging resistant stems and non-shattering seed cases so in cut-flower use they do not shed seeds. They performed well at Reiman Gardens and in several years of plantings at the NCRPIS; I hope that they will be used by the horticultural industry. More information is on-line at:
http://www.ag.iastate.edu/centers/cad/amaranth.html

I am continuing to enhance grain amaranths for reduced lodging and reduced shattering. Eleven breeding lines were field planted and seven will be advanced because of desirable performance. If possible these elite lines will be planted for additional evaluation by three collaborators in other mid-western states.

We are collaborating with Dr. David Baltensperger of the University of Nebraska in an attempt to develop new proso millet (*Panicum miliaceum)* cultivars that are genetically quite different from existing cultivars. The existing cultivars used in Nebraska are from a narrow genetic base. Hybrid seeds of proso millet were produced from a cross between PI 583347 X PI 269954. F_3 generation seeds were shipped to Dr. Baltensperger for evaluation and selection in his environment.

Charlie Block and I continue our work on *Phomopsis amaranthicola* resistance in *Amaranthus tricolor*. Charlie and his group conducted evaluations in their fall 2005 greenhouse. I am

attempting to pyramid the resistance derived from PI 599683 and PI 608761 in breeding crosses. Whether they have different pyramidable resistance genes is to be determined.

I was unsuccessful again in an attempt to cross disease resistant types of *A. tricolor* with the chlorophyll deficient ornamental types. The F1 plant had few flowers and the flowers were malformed and not fertile.

Publications and presentations:
Brenner, D.M., 2005. Amaranthus. p. 99. In Guinness World Records 2006. 2006 ed. Guinness World Records, Ltd., England (World Record tallest amaranth grown at the NCRPIS in 2004, 4.61 m)

Brenner, D.M., 2005. Methods for *Melilotus* germplasm regeneration. Plant Genetic Resources Newsletter 141:51-55.

Brenner, D.M., Dealing with Dormant Seeds. Oral presentation. December 5-6, 2005. USDA, ARS National Plant Germplasm System
Curator Workshop. Chicago, IL.

Brenner, D.M., Enhancement Breeding of Amaranths for Improved Grain, Vegetable, and Ornamental Use. Oral presentation. August 9-10, 2005. Amaranth Institute Biannual Meeting. Bowling Green, KY.

David M. Brenner, Tomas Ayala-Silva, Barbara Hellier, Kim E. Hummer, Maria Jenderek, Laura Fredrick Marek, J. B. Morris, Randall Nelson, K. R. Reitsma, Larry D. Robertson, Susan M. Stieve, Ed W. Stover, and Mark P. Widrlechner. 2005. Genetic Resources of Omega-3 Fatty Acid Crops. *In* 2005 Agronomy Abstracts. ASA, Madison, WI. Poster presentation at the American Society of Agronomy meeting November 6-10, 2005, Salt Lake City, UT.

Costea, M., D.M. Brenner, F.J. Tardif, Y.F.Tan, and M. Sun. 2005 Delinitation of *Amaranthus cruentus* L. and *Amaranthus caudatus* L. using micromorphology and AFLP analysis: an application in germplasm identification. Genetic Resources and Crop Evolution (advance publication on-line)

I prepared written Progress Reports for the Crop Germplasm Committees: Clover and Special Purpose Legumes, Forage and Turf Grass, Leafy Vegetable, and New Crops.

I helped organize a continuing education tour of the Reiman Gardens greenhouse facilities for NCRPIS staff on March 31, 2005. About twenty people from our staff attended.

Plans for 2006:
There are substantial taxonomic problems in the millet collections. I plan to develop my taxonomic expertise for the millets and conduct a series of taxonomic comparison grow-outs, utilizing expertise from collaborators.

Acknowledgments:
Amber Bartels, Alex Fales, Andrew Martin, and Staci Schmit worked with us as part time student help.

Mr. Samuel Flomo continues to expand in his role as the Technician. He gives me opportunity to concentrate on problem areas in the germplasm collection.

Some research publications derived directly from use of our germplasm and associated information:

Atul, Bhargava, Shukla Sudhir, and Ohri Deepak 2005. Analysis of genotype x environment interaction for grain yield in *Chenopodium* spp.
Czech Journal of Genetics and Plant Breeding 41:64-72.

Coles, N.D., C.E. Coleman, S.A. Christensen, E.N. Jellen, M.R. Stevens, A. Bonifacio, J.A. Rojas-Beltran, D.J. Fairbanks, and P.J. Maughan 2005. Development and use of an expressed sequenced tag library in quinoa (*Chenopodium quinoa* Willd.) for the discovery of single nucleotide polymorphisms. Plant Science 168:439-447.

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F. Horticulture (M.P. Widrlechner, J. Carstens)

Germplasm Collections:

Acquisition:

During 2005, we entered information about 29 new accessions of ornamentals into the GRIN database (Table 1); 52 additional ornamental accessions and a mint-family accessions obtained in 2005 will be accessioned early in 2006. The largest groups of new acquisitions included collections made through exchanges from the Republic of Georgia and the Palestinian Territories, as well various tree and shrub clones obtained for possible testing in the NC7 Ornamental Trials. In addition, we were able to re-obtain samples of two accessions that were pending inactivation due to past losses.

Maintenance:

Field plantings of trees and shrubs received special attention for maintenance in 2005, with large-scale transplanting of many accessions that had been held in containers into clean-cultivated nursery rows.

Availability: During 2005, approximately 43% of the ornamental collections and 56% of the mint-family plants were available for distribution (Table 1), figures slightly below those reported in 2004 (43 and 59%).

Back-up: Approximately 32% of the ornamental collections and 59% of the mint-family plants are duplicated at NCGRP (Table 2), figures slight above those reported in 2004 (31% and 59%).

Regeneration: Regeneration efforts intensified somewhat in 2005, with expansion of a three-year cage field established for shrubs, annuals and tall, biennial Malvaceae in 2004, and the establishment of a new, three-year cage field focusing on *Potentilla*, Lamiaceae, and Malvaceae in 2005. The harvests listed in Table 2 include 38 successful cage increases and 41 woody-ornamental seed increases. There were also 56 accessions of woody plants established from seeds and 63 accessions vegetatively re-propagated.

Viability Testing: In 2005, 54 ornamental and 4 mint-family accessions were tested for germination (Table 2). Viability tests for 2005 regeneration lots will begin in January 2006 and will be included in the statistics for the 2006 Annual Report.

Distribution:

As summarized below (and in Tables 3A and 3B), requests for accessions of ornamental germplasm decreased in 2005 below levels in the past five years, both in relation to the number of recipients and the number of items shipped**.** The 241 "order items" included all the distributions for the NC7 Trials (described in the following section), along with 14 plants, 123 cuttings, 83 budwood sticks, and 207 seed packets, distributed to fulfill external requests for ornamental plant germplasm.

This group encompassed 54 genera; those most in demand were *Salix* (103 cuttings), *Fraxinus* (80 budwood sticks and 1 seed packet), *Cornus* (15 packets and 2 plants), *Alcea* (14 packets) and *Malva* (14 packets).

In contrast to decreased demand for the ornamental collections, 59 seed packets were distributed of mint-family germplasm, the most since 1998.

Historical Summary of Distribution Activity:
Note: In the summer of 2004, about 240 accessions of *Echinacea* and *Hypericum* were transferred to Joe-Ann McCoy for curation. In addition, about 500 accessions of herbaceous ornamental germplasm representing 23 genera once part of this project were transferred to the Ornamental Plant Germplasm Center (OPGC) in Columbus, Ohio for maintenance in 2002. Statistics presented in the summary tables at the end of this Annual Report exclude activity in the transferred accessions. However, for comparative purposes, statistics reported in the historical distribution table (below) do include activity related to the transferred accessions conducted prior to their transfer.

Crop	Year	No. of Orders	No. of Recipients	No. of Items Distributed	No. of Accessions Distributed
Ornamentals	01	94	85	671	365
	02	103	89	779	361
	03	108	91	883	320
	04[1]	87	81	361	297
	05[1]	58	53	241	187
Mint Family	01	5	5	42	42
	02	4	4	22	19
	03	9	9	45	39
	04[1]	17	16	45	37
	05[1]	17	16	59	38

[1] Includes external distributions only.

Characterization/taxonomy:
All the herbaceous ornamentals in annual cage field and many of the tree and shrub accessions being regenerated were checked to verify identifications. In all, 10 ornamental and six mint-family accessions were re-identified. During 2005, Jeff Carstens captured images of 62 ornamental and mint-family accessions for our local database (Table 4). These are named following our standard protocol and will be loaded to GRIN early in 2006, by using the new mass-loading system for images, currently being developed by Pete Cyr.

Evaluation:
An extensive collection of evaluation data was received from NC-7 trial-site cooperators and has been loaded into our Access database. Selected data were also summarized and prepared for loading to our Internet database (described further in the section "Coordination of the NC-7 Regional Ornamental Trials").

Enhancement:
There was no major progress to report with enhancement activities in 2005.

Coordination of the NC-7 Regional Ornamental Trials:
Plant Distribution - In 2005, Mark Widrlechner and Jeff Carstens distributed 208 plants of five accessions to 18 sites for long-term evaluation, with an additional 77 plants of these accessions provided to 11 public gardens. As part of that process, during May, Mark Widrlechner and Jeff Carstens delivered plants and met with cooperators in South Dakota, North Dakota, and Minnesota.

Computer-generated, "One-, Five-, and Ten-year Performance Report" forms were distributed to trial-site cooperators this summer, and all data returned by cooperators in 2004 were recorded in Access in 2005. Evaluation data submitted by the cooperators covering distributions made from 1991 through 1993 were summarized and posted in Public GRIN in a new format utilizing Accession Actions.

An extensive collection of reports on the evaluation of NC7 Trial plants was published from the 1960s until about 1980. These reports are not widely available. During 2002, Kyle Cavanaugh scanned these reports and created .pdf files. These reports have been indexed and will also be placed on our website in 2006.

Two updates were emailed or sent to trial cooperators in 2005 to inform them about recent developments in the testing program, plans for future distributions, and ongoing efforts to regenerate *Potentilla* germplasm.

Germplasm activities in crops other than those curated:
Throughout 2005, Mark Widrlechner actively participated in a university-industry-ARS collaboration to guide the development of the Ornamental Plant Germplasm Center (OPGC), in Columbus, OH, and facilitate its integration within the National Plant Germplasm System. He serves as the Agency's representative to administer a Specific Cooperative Agreement (SCA) between ARS and The Ohio State University to fund the OPGC. In 2005, he helped assemble an external team to conduct a Project Planning Session at the OPGC early in 2006. This team will make recommendations about future plans for the Center, establishing new goals and milestones.

In 2002, Iowa State University and the University of Iowa were awarded a five-year grant from the National Institutes of Health (NIH) establishing a Center for Research on Botanical Dietary Supplements to study variation and bioactivity in *Echinacea* and *Hypericum*. Mark Widrlechner has continued his involvement with the Center by overseeing a subcontract to ARS, which supports the curation of the Station's *Echinacea* and *Hypericum* germplasm collections by Joe-Ann McCoy and the distribution of that germplasm so it can be evaluated for chemical composition, genetic diversity, and bioactivity. In 2005, work began to develop a proposal to NIH to request funding to extend Center activities for the period, 2007-2010.

During 2005, Mark Widrlechner was involved with a number of other collaborative germplasm activities including:

1. a *Coriandrum* germplasm evaluation project being conducted with Pedro Lopez, a doctoral candidate at Iowa State University, who focused on data analysis, vouchering samples, and developing AFLP-marker systems to study patterns of genetic diversity, and with Terry Isbell, National Center for Agricultural Utilization Research, Peoria, IL, who evaluated fatty-acid composition in its seed oils;

2. completion of a comprehensive literature review on cucurbit germplasm for a chapter in the series "Genetic Resources, Chromosome Engineering, and Crop Improvement," with Aleš Lebeda, Palacký University, Czech Republic, Jack Staub, USDA-ARS, Madison, WI, et al.;

3. completion of a project that evaluated variation in seed dormancy in wild and cultivated populations of *Echinacea purpurea* with Luping Qu et al. of Gaia Herbs, Inc., Brevard, NC, which was published in HortScience;

4. assistance in the completion of a paper which was accepted by Crop Science titled, "Demand for Genetic Resources and the U.S. National Plant Germplasm System," based on research initially conducted by Kelly Day Rubenstein (USDA-ERS) and Melinda Smale (IPGRI);

5. assistance in the completion of a manuscript on the use of liquid-nitrogen to control chalcid wasp infestations in *Coriandrum* seeds, involving David Kovach, Sharon McClurg, David Brenner, and Candice Gardner to be submitted to Seed Science & Technology early in 2006; and

6. completion of draft national policies on germplasm acquisition and distribution through service in the Plant Germplasm Operations Committee.

Research products:
The project to evaluate variation in seed dormancy in *Echinacea purpurea* (point 3 above) revealed evidence to support the hypothesis that seed dormancy in this species has been greatly reduced during repeated cycles of cultivation for ornamental and medicinal uses, a common step in crop domestication. Findings from this project are also useful for commercial growers who need to ensure consistent seed germination and stand establishment.

The successful completion of a project to develop a system to control *Systole* chalcids in *Coriandrum* seeds (point 5 above) will benefit the conservation and safe distribution of coriander germplasm by preventing future spread of this insect pest and potentially lead to the development of similar systems to control other seed-borne insects. See the "Seed Research and Computer Application Development" Section for more details.

Mark Widrlechner's other research and training activities:
Collaboration continued with Welby Smith of the Minnesota Department of Natural Resources leading to the completion of the treatment of *Rubus* for a new book on the woody plants of Minnesota.

Collaborations also continued on the development of models to predict the risk of naturalization of non-native woody plants. In 2005, Mark Widrlechner presented an overview of this work to the 2005 Annual Meeting of the American Association of Botanical Gardens & Arboreta in Chicago. During 2005, significant progress was made on the assembly of data describing an extensive set of naturalizing and non-naturalizing woody plants from the Chicago region, in an effort to validate risk-assessment models developed from data collected in Iowa. This work is being conducted in concert with Peter Bristol and Kristen Kordecki at the Chicago Botanic Garden, funded through a Specific Cooperative Agreement.

In 2005, Mark Widrlechner continued his service as chair of a national Technical Review Committee that provides technical direction and oversight to an ARS project to update the USDA Plant Hardiness Zone Map by using the best available technologies and to make the next version of the map accessible via the Internet.

In March, 2005, Mark Widrlechner participated in the ARS Congressional Briefing Conference in Washington, DC, where he learned about the Congressional budgeting process. In June, he completed reviewer training for the North American Plant Collection Consortium. He represented the NPGS in September at "New Roots for the 21st Century – US-Russia Botanical Conference," a meeting in Chambersburg, PA sponsored by the US National Arboretum and the US Fish & Wildlife Service and there helped plan future research collaborations with Russian and American botanists. And in December, he presented information on germplasm inactivation protocols to the NPGS Curator Workshop in Rosemont, IL and gained considerable new information about curatorial management by attending.

Other Horticultural project-training and staff-development activities:
In 2005, Mark Widrlechner and Jeff Carstens completed training sessions to satisfy continuing education requirements for tractor safety, hearing safety, ARS's Environmental Management System, IT security, and state pesticide applicator certification. Jeff also attended the NPGS Curator Workshop in Rosemont, IL and training for hazardous waste generators and for respirator certification.

Communications Activities:
Manuscript and Proposal Review:
Mark Widrlechner continued his service on the Editorial Review Boards of Genetic Resources and Crop Evolution and the Journal of the American Rhododendron Society. He also served as a peer reviewer for manuscripts submitted to HortTechnology, Journal of Herbs, Spices and Medicinal Plants, Maydica, and Plant Cell Reports, and as an internal reviewer for ten papers prior to journal submission. In addition, he reviewed a grant proposal for the U.S. Civilian R&D Foundation.

Posters, Presentations and Seminars:
Brenner, David M., Tomas Ayala-Silva, Barbara Hellier, Kim E. Hummer, Maria Jenderek, Laura Fredrick Marek, J. B. Morris, Randall Nelson, K. R. Reitsma, Larry D. Robertson, Susan M. Stieve, Ed W. Stover, and Mark P. Widrlechner. 2005. Genetic resources of omega-3 fatty acid crops. Poster presented to the The ASA-CSSA-SSSA International Annual Meetings (November 6-10, 2005), Salt Lake City, UT. Abstract published on CD: Abstracts 2005 International Annual Meetings.

McCoy, Joe-Ann, Mark Widrlechner, and Jeff Carstens. 2005. A comprehensive *Echinacea* germplasm collection located at the North Central Regional Plant Introduction Station, Ames, Iowa. HortScience 40: 1063 (Poster Abstract for 2005 ASHS Annual Conference).

Widrlechner, Mark P. 2005. Invasive risk assessment: A regional perspective. Invited talk in session, "Invasive Risk Assessment: Methods to Evaluate New Taxa for Invasive Characteristics," AABGA Annual Conference, Chicago, 2 July.

Publications which appeared in print in 2005:
Qu, Luping, Xiping Wang, Ying Chen, Richard Scalzo, Mark P. Widrlechner, Jeanine M. Davis, and James F. Hancock. 2005. Commercial seed lots exhibit reduced seed dormancy in comparison to wild seed lots of *Echinacea purpurea*. HortScience 40: 1843-1845.

Widrlechner, Mark P. 2005. Germplasm acquisition from conception to products. (Introduction to Workshop Proceedings). HortScience 40: 296.

Departmental Activities:
Mark Widrlechner continued as an active member of the Crop Seeds Committee and the Plant Breeding and Genetics Advisory Panel of the Agronomy Department at Iowa State University. He also served on Agronomy Department's Greenhouse & Growth Chamber Committee and the faculty of the Horticulture Department. He continued to serve as Co-major Professor for Pedro Lopez, a Ph.D. candidate in Plant Breeding, and on the Program of Study (POS) Committees for two Ph.D. candidates, one in Plant Breeding and the other in Horticulture. He served on the POS Committee for Evelyn Ortiz-Perez, who successfully completed her Ph.D. in Plant Breeding in 2005, and became a member of the POS Committee for a Ph.D. candidate in the Interdepartmental Plant Physiology Program.

Conclusions and Plans for 2006:
Curation:
Curation efforts in 2005 focused on increasing levels of activity in caged seed multiplication and increasing the efficiency of our handling of longer-term tree and shrub plantings. The completion of Joe-Ann McCoy's first full field season also provided many opportunities for mentoring on the fine points of collection curation.

Looking ahead to 2006, we have large numbers of accessions of herbaceous perennials and shrubs in place for significant expansion of our caged-seed regeneration effort over past years. In addition, we will fill in blank spots with annuals, such as *Calendula* and *Glebionis*, to maximize use of our cages.

For the tree genera, *Alnus* and *Betula*, Jeff Carstens continues to learn more about the development of controlled-pollination systems for seed production. And given the serious threat caused by the introduction of Emerald Ash Borer to the North Central Region, additional attention will be given to the collection and conservation and regeneration of *Fraxinus* germplasm through collaboration with David Ellis, Kevin Conrad, and Ned Garvey.

To help capture valuable information about our accessions, we have begun developing descriptor lists for *Calendula*, a crop with ornamental, medicinal, and industrial uses, and for *Potentilla*. Draft lists will be tested during the 2006 field season and then brought to the New Crops and Herbaceous Ornamental CGCs for their review and approval.

Research:
Considerable progress was made on a wide range of research projects during the past year as outlined above.

Research efforts for the coming year will focus on:

1. validating recently developed risk-assessment models for the invasiveness of non-native woody plants in the Midwest with data being collected from the Chicago region through the support of a Specific Cooperative Agreement with the Chicago Botanic Garden (attempts are also underway to secure additional external support for this project);

2. coordinating technical advice and relaying appropriate research information in support of efforts by Robert Webster, USDA-ARS, Beltsville, MD, to revise the USDA Plant Hardiness Zone Map;

3. completing and submitting a study for publication that uses long-term germplasm viability records and distribution histories to estimate target quantities for seed regeneration;

4. following-up on initial experiments conducted by Leigh Towill to develop practical protocols with Gayle Volk, NCGRP, Fort Collins, CO, for the routine cryogenic storage and plant recovery of *Salix* clones; and

5. providing guidance to Pedro Lopez as he completes his studies on patterns of genetic and phenotypic diversity in *Coriandrum*.

Staff Development:
Plans for staff development for 2006 will focus on training experiences for Jeff Carstens, which are likely to include attendance at the Eastern Region Annual Meeting of the International Plant Propagators' Society, the Iowa Shade Tree Short Course, visits to NC7 Ornamental Trial sites, local nurseries, and safety training.

G. Maize Curation (M. Millard, M. Lively, D. Losure)

Equipment:
Equipment acquisition for the maize program was minimal in 2005. An HP iPAQ hx4700 and a Palm Zire 72 were acquired during 2005 for comparison of field data acquisition features. At this time, the HP is the desired device as it is more visible in daylight than the Palm and it works more closely with Microsoft software. Data-On-The-Run version 4.07 was used on the HP. A form was created for field plant measurements on maize increases and fed directly into Microsoft Access for final data processing and uploading to GRIN. A Barcoda Bluetooth barcode scanner was purchased for scanning barcodes from tags into the data acquisition program. It was determined that this type of scanner is not appropriate for the Tyvek labels the maize project uses because scanning requires a smoother label and contact with the label. While laser scanners can scan the rough resolution of the Tyvek barcodes even when dirty, the Barcoda often times fails to read these codes and dirt will wear on the scanner surface.

Personnel:
Last year's NCRPIS report raved about reaching 2002 staffing levels, setting the stage for disappointing staffing developments in 2005. A considerable number of staff changes occurred on the Maize curatorial team. Ms. Mindy Weishaar resigned in April, 2005 to join Pioneer Hi-Bred International, Inc. She worked as the federal maize technician for seven months. We wish her well in her new career. Ms. Gaylan Crim resigned in June, 2005 to pursue other interests. Ms. Crim was one of the most capable individuals at the North Central Regional Plant Introduction Station (NCRPIS) and will be sorely missed. She worked as the state maize technician for over 6½ years.

Re-staffing began when Mark J. Millard, the maize curator, switched affiliations from the state curator position funded under the NC7 project to a federal GS-11 geneticist position after a successful application-interview process. The federal geneticist position became available with the advent of the USDA FY 2006 in October. The vacant federal maize technician position was filled by Mr. Matt Lively who had previously worked in the corn breeding program of Garst Seeds. Matt returned to the NCRPIS in November 2006. He had once worked as a student on the maize project and also worked maintaining bees for the entomology project. Welcome back Matt! The vacant state maize technician position was filled by Mr. David Losure. David is finishing a master's thesis in ecology and evolutionary biology at Iowa State where he was also a teaching assistant. He began work after the start of calendar year 2006.

The vacant state maize curator position will be reworked in 2006 and opened when funding permits. Increased Hatch or ARS funding prospects at this time appear unlikely and an increase in staffing may have to wait for some time. Staffing remains at levels which were first achieved in

1998; a permanent staff of four would be very advantageous. New challenges for the program will need to be met with the renewed energy that a new staff often brings to a project.

Research Progress:

The following paper was published in 2005:

> Desjardins, A.E., R.D. Plattner, R.J. Stessman, S.P. McCormick and M.J. Millard. 2005. Identification and heritability of fumonisin insensitivity in *Zea mays*. Phytochemistry 66(20):2474-2480.

This paper indicates that the accession Tama Flint (PI 217411) may have some seedling tolerance to fumonisin while other related accessions by race as well as many other unrelated accessions do not.

The thesis project on the races of maize in the Southwestern U.S. continues; Ms. Lindsay Werth is studying the landraces of maize native to the U.S. Southwest with the aim at describing the races found there. Dr. Debra Muenchrath is the major professor for these studies, and Dr. Candice Gardner, the NCRPIS Research Leader, is also involved in the project. A large observation nursery was grown successfully at the New Mexico Agricultural Experiment Station in Farmington in 2004 and 2005, yielding many specimen or "show" ears for the project. A location was also grown in Ames in 2004.

The CD's of the Races of Maize Monograph Collection have been completed and will be sent out upon request to the curator, Mr. Mark J. Millard, mjmillar@iastate.edu. This version includes the Spanish version of the Races of Maize for Mexico, Raza de Maiz en México, which was a challenge to complete. We found it difficult to obtain a pristine version. Dr. Major Goodman was able to locate and obtain over the Internet a leather-bound copy of Dr. Wellhausen's. The NCRPIS staff felt that this copy's condition was too good to disassemble as had been done with the other paperback editions. The book was scanned manually by NCRPIS staffer Ms. Cris Nass using the Iowa State Library's book scanner. This was a slow process compared to the document feeder process used for the other volumes. After the book was imaged, optical character recognition (OCR) was attempted, but it was found that the scanning of the book changed fonts enough to make OCR much less precise than had been done on earlier books. Therefore, a copy of the book provided by Dr. Wilfredo Salhuana that had been heavily annotated was scanned and OCR'ed. Pages that were heavily annotated were exchanged with pages from Wellhausen's book. Even then, some pages had to be manually touched up with Adobe Photoshop for accurate OCR and for appearance. Several at the NCRPIS helped with this project, but special thanks to Pete Cyr for helping with technical details. We will work over the next few months to place these documents on the Internet. We will also add other Spanish versions so graciously provided by Dr. Major Goodman and Dr. Wilfredo Salhuana as permissions are obtained and as time permits.

Dr. Ed Buckler agreed to do quality assurance using SSR markers to verify the quality of 89 accessions. These accessions are inbred lines in the Goodman diversity set that had been previously grown to large quantities by the NCRPIS. We were seeking to determine whether previously regenerated samples compare well with Goodman samples in order to save resources that would be utilized by regenerating these again from Goodman seed. Dr. Buckler returned summary data indicating that only 5 of the 89 may need to be grown from Dr. Goodman's seed. The differences appeared to be due to a difference in seed source and not to contamination issues. This will save increasing 84 of the 300 lines, a significant cost savings. The CGC recommended, at the suggestion of the maize curator, that two versions of these lines be maintained. Those in the collection previous to Dr. Goodman's deposit will maintain the original line designation. Those in the Buckler-Goodman set will be designated: P39 (Goodman), IA2132 (Goodman), N7A (Goodman), CI91 (Goodman), CI7 (Goodman).

Acquisition:
There were 75 new accessions incorporated into the collection in 2005 as seen in Table 1. Thirty-four of these were expired Plant Variety Protected (PVP) accessions. Twenty-one were U.S. Crop Science Registered (CSR) accessions. The remainder of the accessions received were CSR inbreds from Nigeria which were regenerated in quarantine on St. Croix.

The NCRPIS has imaged all of the Goodman Racial Collection of Maize received in June, 2004. The collection consisted of about 1,800 accessions. There were 2,110 images of seed taken. Crosschecking with the collection has begun, but was delayed due to the staff changes in 2005. We plan to fully incorporate the inventory into the collection in 2006 and continue with the action plan given to the Maize CGC in 2004 and reported in the 2004 NCRPIS annual report.

Regeneration:
There were 130 accessions attempted in FY '05 in Ames compared to 317 in FY '04 (Table 2). This nursery was greatly reduced in size due to the loss of the only two maize techs before planting. A second difficulty was that the student help were all new except for one individual. Emphasis was placed on increasing the expired PVPs and early inbreds to make the regeneration season as short as possible. This worked well and harvest was completed almost three weeks earlier than most years. One difficulty the season presented was a dry period in the middle of the pollinations on the earliest accessions. They were irrigated, but their production was affected. On the whole, seed production was a little better than average.

One annual teosinte, the only active accession in NPGS of *Zea nicaraguensis* was attempted in FY '05. The accession was planted in November 2004 and failed to flower under short day conditions in the greenhouse of less then 12 hours of light. The plants continued to grow into the rafters at 15 feet or more. Cuttings were made during the next spring and summer from the tops of the main stalk and tillers below the first node fully emerged below the top of the stalk. These tops rooted without assistance in a gallon of water and were transplanted to soil. They are now flowering in the greenhouse. Visiting scientist Ana Gulbani's efforts were instrumental in the success of this project.

Seed from 2 accessions of *Zea perennis* were harvested from four accessions grown in the Ames greenhouse during FY '05. These accessions can be maintained clonally indefinitely. As seed is harvested, the clones are discarded.

Fifty accessions were sent to the Golden Harvest facility near Ponce, Puerto Rico for increase. These have been received in FY '05 in good condition.

Two groups of 50 accessions were sent to Carlos Hernandez, owner/manager of CH Farm Services, Inc., for increase in FY '05. One of these has been grown and increased. The second nursery was delayed in planting due to above average rains near Ponce and will be received in FY '06.

Fifty-four accessions were sent to St. Croix for increase in FY '05 for increase and have been received.

Sixty-seven accessions were sent to ICIA on Puerto Rico near Ponce in FY 05'. These were pollinated by the maize curator and federal maize tech with the assistance of the ICIA staff. These consisted of several of the Goodman-Buckler reference lines more adapted to the tropics. This nursery has been processed and made available

Fifty-five accessions were received from Syngenta at their Hawaii facility. These were accessions grown from Major Goodman's racial collection. We received the excess seed on the ear for backup of what we had received from Dr. Goodman.

Ten accessions were grown by Syngenta as tropical increases by Mr. Mark Lauchner. Thanks to Syngenta for both of these increases.

Maintenance:
Table 1 indicates that accession availability slightly increased to 66% an increase of 235 available accessions over last year. The 436 accessions made available in 2005 were countered by 201 accessions made unavailable due to depleted supply or reduced viability. These numbers do not include the Goodman Maize Race collections of 1,800+ accessions as these have not yet been completely incorporated into the collection statistics. This means that the percent of the collection available will go down significantly next year.

In 2005, 1332 accessions, or 7%, of the collection were tested for viability compared to 970 or 5% in 2004. Eleven percent of available accessions were germinated. Viability testing detected 7 accessions (.5% of those tested) that were less than 50% viable and taken off distribution. This compares with 7 accessions (.7%) in 2004 and 33 (3.7%) in 2003. Distribution was raised from the standard 100 kernels to 200 kernels on 86 accessions that have plenty of seed, but whose viability was between 51% and 84%.

Distribution:
Table 5 shows that maize packet distributions were about the same in 2005, but both the number of cooperators receiving seed and the number of orders was up significantly. The expired PVP inbred lines continued to be a popular distribution. There were 77 accessions in this group by the end of 2005 and 104 of the orders (27% of all maize orders) and 1164 packets (26% of all maize packets).

Crop	Calendar Year	No. of Orders	No. of Recipients	No. of Items Distributed	No. of Accessions Distributed
Maize relatives	2001	7	7	13	7
	2002	5	5	16	7
	2003	7	7	22	8
	2004	8	8	11	6
	2005	7	7	11	6
Average		**7**	**7**	**15**	**7**

Maize	2001	270	198	2637	1606
	2002	399	279	4714	2511
	2003	226	178	2298	1475
	2004	335	241	4973	2617
	2005	380	274	4422	1826
Average		**322**	**234**	**3809**	**2007**

Characterization:
There were 8,310 data points relating to 14 ear descriptors on 478 accessions loaded into GRIN in 2005. This compares with 7,480 data points relating to 14 ear descriptors on 335 accessions loaded into GRIN in 2004. There were 13,062 data points on 980 accessions in 2003. The ear description should pick up even more in 2006 since the staffing is up as long as regenerations increase.

We obtained 7121 images on 3556 accessions in the collection compared to 3035 images on 806 accessions in 2004 and 4,512 images on 1602 accessions in 2003. There is at least one digital image on 16,324 accessions or 89% of the collection on the books. Not included in these figures are the 2110 images taken on the 1800+ accessions from the Goodman racial collection which has not yet been incorporated into the collection. When this is completed in 2006 it is possible that the entire collection will have at least one image available to the maize staff. This will be a significant benchmark.

Evaluation:
The maize disease resistance-screening program continued in 2005.

Three hundred accessions were sent to Dr. S. Moore at Louisiana State University for aflatoxin production and Aspergillus ear rot resistance trails. These were topcrossed with Mo17 for trials in later years. Eight hundred accessions have been sent in previous years and their topcross tested. The 11 accessions in the table have been identified as possibly supporting low Aflatoxin producers in combination with Mo17. They will undergo further testing.

Accession	ID	Origin	Type
NSL 75956	NIGERIAN COMPOSITE B	Nigeria	Population
PI 490411	Manio	Mali	Population
PI 280061	Lancaster Surecrop	Ohio, United States	Population
PI 218189	P 69	Arizona, United States	Population
PI 483902	Haiti 33	Haiti	Population
PI 484036	Saint Croix 1	St. Croix, Virgin Islands (U.S.)	Population
PI 485481	Dominican Republic 309	Dominican Republic	Population
PI 506253	Tzi 18	Oyo, Nigeria	Inbred
Ames 22761	Tx81	Texas, United States	Inbred
PI 619430	Tx807	Texas, United States	Inbred
PI 595535	CML 43	Federal District, Mexico	Inbred

Dr. W. Dolezal with Pioneer Hi-Bred International, Inc. planted 250 accessions in Johnston, Iowa in 2005. Northern leaf blight resistance was observed under artificially infested field conditions with a high incidence of the disease. Thirty-nine accessions were identified as rating a 7 or 8 on the Pioneer 1-9 scale, with 9 being most resistant. These included several North Carolina lines and three GEM accessions. He hosted a tour of the nursery at Pioneer for the NCRPIS maize curator, GEM coordinator, the pathologist, and the research leader in Johnston. Additionally he hosted the NCRPIS maize curator and GEM coordinator at the Pioneer station at Woodland, California where they conduct fusarium ear rot screening. Good resistance was demonstrated by GEM accessions that have part of their origin in NCRPIS maize accessions. Thanks to Pioneer and to Dr. Dolezal for their generosity in this work.

Dr. C. Block of the NCRPIS evaluated 380 accessions for Stewart's wilt resistance. The majority of this nursery consisted of inbred lines including the expired PVPs. New GEM accessions were also screened.

Communication:
Many groups who tour the farm observe the maize imaging project as part of their learning experience. This has been a popular demonstration over the past few years with visitors, and has increased awareness of image availability in GRIN. Especially notable this year was a visit by the ARS Administrator's Council members who had many good questions concerning the project. The demonstration was enhanced by large poster size images selected by David Kovach, Candice Gardner and Larry Lockhart from previously taken images in the same manner as all the maize kernel images. Dave Kovach printed large poster size images of each using the NCRPIS large format printer.

Plans for 2006:
Acquisition:
We are planning to complete the inventory and image documentation of the Goodman Maize Race Collection. We will then complete a prioritization and start the acquisition and regeneration process.

More GEM accessions will be released for distribution by the NCRPIS in fiscal '06.

The NCRPIS, NCGRP, CIMMYT, and Dr. Wilfredo Salhuana will continue a project to pull together a consolidated list of the holdings of the various countries' collections and the CIMMYT and U.S. collections to evaluate material that may be at risk.

Regeneration:
Funding will support tropical maize regenerations of 150 to 200 accessions per year by private sector nursery providers to NCRPIS specifications.

Regenerations in Ames will be maintained at 250-300 accessions annually

Quarantine regenerations on St. Croix will continue at the 30-50 accession level during 2006.

Maintenance:
The maize staff will be reviewing their processes looking for changes that will gain productivity.

Dr. Buckler has agreed to quality screen with SSRs a large group of increases of the Goodman-Buckler diversity set the winter of 2005-06.

The NCRPIS, NCGRP, CIMMYT, and Dr. Wilfredo Salhuana will work on a continuing project to develop and cross reference a consolidated list of the holdings of the various L.A. countries' maize germplasm collections, the CIMMYT collection, and U.S. collections in order to identify materials that may be at imminent risk of loss.

Viability tests will be increased in 2006 to maintain the maize testing schedule to provide timely data for regeneration priorities.

Evaluation:
I will attempt to augment the collection of images currently on GRIN of 4,500 accessions with images of an additional 10,000 accessions in FY '06. These images have already been taken, but will be loaded with the new GRIN loader software developed by the NCRPIS.

The maize curator will continue to work with the NCRPIS pathologist and interested private and public pathologists to systematically obtain data on maize pathogen resistance in the collection. Additional evaluation information will result in more effective use of accessions in the future.

The project will continue planting observation plots to obtain maturity data. Additionally, older data from field book sources will be gleaned for inclusion into GRIN. Maturity data is one of the most important criteria for determining selection of accessions which meet researchers' objectives and allows them to plan their work.

We will continue to load existing molecular marker information and frequencies on collections held at the NCRPIS. After finishing the N.C. State isozyme data, we will add SSR data obtained at NCRPIS. Dr. Laura Marek, the oilseeds curator has expertise in this area and will assist in refining the current design for the molecular marker schema in GRIN.

Other Items:
We plan on posting the digital compilation of the Races of Maize Monograph Collection to the Internet in FY '06.

H. Medicinal Plants (J. McCoy)

Germplasm Collections:
Perennial crop collections currently curated by the Medicinal project include *Echinacea (prairie coneflower), Hypericum (St. John's wort), Actaea racemosa (black cohosh),* and various miscellaneous species (NC7 Medicinals).

Acquisition:
During 2005, we received and/or collected 59 new accessions of medicinal species representing 16% of the current collection (Table 1). The collection currently consists of 364 accessions, although the annual statistics imply 305 as passport data entry is currently in process.

Collection trips:
In collaboration with curator Laura Marek, a short collection trip was conducted June 10-15 to Oregon following the annual PGOC Conference. Eight accessions were collected and negotiations were arranged with a researcher to secure future accessions, which have been partially received. A second collection trip was coordinated by Joe-ann McCoy in September, 2005 in collaboration with botanists, Dr. Chick Gaddy, Patrick McMillan, and Dr. Richard Porcher which resulted in 20 new accessions. This collection trip covered portions of western North Carolina and the coastal plains and piedmont sections of South Carolina. The main emphasis of the trip was the collection of native *Hypericum* germplasm; the Federally endangered *Echinacea laevigata,* was also collected.

Availability and Backup:
Fifty percent of the NC7 medicinal accessions are currently available (Table 1.). Fifty accessions have been harvested and 42 made available in 2005. Fifty four accessions have been backed up in 2005, with a total of 152 accessions now backed up in Fort Collins, representing 50% of the total collection (Table 2).

Regeneration and Maintenance:
The majority of the growing season was spent maintaining 78 field cages. Seeds from 50 accessions of *Echinacea* and *Hypericum*, which were regenerated in field cages, were harvested, and are currently being processed and stored (Table 2). Regeneration efforts focused on new *Hypericum* accessions for a field planting in 2005 which consisted of 42 new field cages. Vegetative and

rhizome samples of these new accessions were also made available to ISU / NIH researchers for both chemical and genetic analyses.

Distribution:
Three hundred and seventy seven items were distributed in 2005; of these, 69% were domestic while 31% were foreign distributions (Table 3A). Along with seed distribution, leaf samples from *Hypericum* and *Echinacea* accessions grown under greenhouse conditions continue to be distributed for molecular-marker analyses to Dr. Jonathan Wendel's lab and to Dr. Eve Wurtele's lab for associated HPLC analysis. Both projects are associated with the ISU NIH Center for Research on Dietary Supplements grant project. *Echinacea* leaf samples were also mailed to Dr. James Price for genome size determination.

Characterization and Taxonomy:
Descriptor data for 43 morphological features were collected from flowering field accessions of *Echinacea*, following the CGC descriptor list protocol as developed by Mark Widrlechner. 6,438 observations for *Echinacea* accessions have been mass loaded into GRIN (Table 4).

Hypericum descriptors are currently in draft format and will be utilized for 2006 flowering accessions. All 2005 blooming accessions were photographed and will be loaded with data in 2006 utilizing standardized file naming protocols.

Pathogen Observations:
Field plantings were monitored weekly during the growing season, for *Colletotrichum* and aster yellows disease symptoms. Both pathogens were present in 2005 field plantings. Positive identification was confirmed for *Colletotrichum gloeosporioides* via spore identification.
A *C. gloeosporioides* seed screening protocol for *Hypericum* accessions continues to be utilized for all new and harvested seed accessions. Forty-three accessions have currently been tested (Table 3B). All germination and pathogen data collected have been entered in the GRIN database.

Joe-Ann McCoy's other research and training activities:
Obtained an IA Pesticide Applicator License; Completed an Introduction to ArcGIS 9 short course; Attended a Curators Workshop in Chicago, IL; Attended the annual PGOC meeting in Pullman, WA.

Regularly attended monthly meetings for NIH cooperators associated with the Center for Research on Botanical Dietary Supplements. In reference to the NIH project, have continued to: 1.)work with Ph.D. students to supply plant material for center projects; 2.) identify and provide commercial *Hypericum* seed to cooperators for field testing and HPLC analysis; 3.) provide relevant journal articles and conference proceedings associated with the project.

Posters / Publication:
McCoy, J., Widrlechner M., and Carstens, J. 2005. A Comprehensive Echinacea Germplasm Collection Located at the NCRPIS, Ames, IA. In Proceedings of the 2005 American Society of Horticultural Science. Las Vegas, NV. July 18-21/2005. 40(4):406. (poster)

McCoy, J. H. 2005. Medicinal Plant Disease List. In Persons, W.S. and J.M. Davis. Growing and Marketing Ginseng, Goldenseal & Other Woodland Medicinals. Bright Mountain Books. p. 423-432. Fairview, NC, 466 p.

Invited Talks:
Black Cohosh (*Actaea racemosa* L.) Research. Growing North Carolina's Natural Product Economy Conference. NC Arboretum, Asheville, NC. 3/4 -5/2005.

Pathogens of Cultivated and Wildcrafted Medicinal Plants. Growing North Carolina's Natural Product Economy Conference. NC Arboretum, Asheville, NC. 3/4 -5/2005.

Annual Meetings:
Have attended annual meetings for 1.) ASHS; 2.) PGOC; and 3.) The International Conference on Quality and Safety Issues Related to Botanicals.

Manuscript Review:
In 2005 eight peer reviewed manuscripts were reviewed prior to submission.

Plans for 2006:
A primary goal for 2006 will be to regenerate and make available a larger portion of the Medicinals collection. Also, voucher specimens will need to be collected for all accessions and labeled and cataloged appropriately. Further acquisition of native medicinal species to increase the Medicinal collection will also be a primary goal. A collection trip to the Republic of Georgia is under discussion with plans to write an exploration proposal in 2006 for the 2007 season. A domestic collection trip will be planned for Fall, 2006 for various medicinal species including *Scutellaria*. *Hypericum* and *Echinacea* descriptor data will be collected for 2006 field accessions.

The curator has been invited to give two talks in 2006 on the status of medicinal plants in the NPGS. (ASHS; ISHS).

A collaboration is being established with Dr. James Price, TA&M to determine ploidy levels via flow cytometry for the *Echinacea* collection and plans are underway to possibly start a similar project with the *Hypericum* collection.

Monthly meetings with post doc researchers and graduate students associated with the NIH Center for Research on Botanical Dietary Supplements will continue to be attended.

Research Impact (recent publications directly associated with accessions):
Hassell, R. L., Dufault, R. J., Phillips, T. 2004. Relationship among seed size, mother plant age and temperature on *Echinacea angustifolia, pallida*, and *purpurea*. Acta Horticulturae. 629:239-243.

Hassell, R. L., Dufault, R. J., Phillips, T., Hale, T. A. 2004. Influence of Temperature Gradients on Pale and Purple Coneflower, Feverfew, and Valerian *Germination. HortTech. 14(3).*

Lata, H., Bedir, E., Khan, I., Moraes, R. M. 2004. Mass Propagation of *Echinacea angustifolia*: A Protocol Refinement Using Shoot Encapsulation and Temporary Immersion Bioreactor. Acta Horticulturae. 629 : 409-414.

McCoy, J.H., Widrlechner, M.P., Carstens, J.D. 2005. A Comprehensive Echinacea *Germplasm Collection Located at the North Central Regional Plant Introduction Station, Ames, Iowa. Hortscience. 40:1063. (Poster)*

Pugh, N., Balachandran, P., Lata, H., Dayan, F., Joshi, V., Bedir, E., Makino, T., Moraes, R., Kahn, I., Pasco, D. 2005. Melanin: dietary mucosal immune modulator from *Echinacea* and other botanical supplements. International Immunopharmacology. 5:637-647.
Qu, L., Wang, X., Chen, Y., Scalzo, R., Widrlechner, M.P., Davis, J.M., Hancock, J.F. 2005. Commercial Seed Lots Exhibit Reduced Seed Dormancy in Comparison to Wild Seed Lots of Echinacea purpurea. *Hortscience. 40(6): 1843-1845.*

Qu, L., Wang, X., Scalzo, R., Widrlechner, M.P., Davis, J.M., Hancock, J.F. 2005. Selection During Cultivation Results in Reduced Seed Dormancy in *Echinacea purpurea* (L.) Moench. Hortscience. 40(6):1843-1845.

Qu, L., Wang, X., Hood, E., Wang, M.H., Scalzo, R. 2004. Chromosome karyotypes of *Echinacea angustifolia* var. *angustifolia* and *E. purpurea*. Hortscience. 39(2): 368-370.

Romero, F. R., Delate, K., Hannapel, D. J. 2005. The Effect of Seed Source, Light during Germination, and Cold-moist Stratification on Seed Germination in Three Species of Echinacea for Organic Production. Hortscience. 40(6): 1751-1754.

Senchina DS, McCann DA, Asp JM, Johnson JA, Cunnick JE, Kaiser MS, Kohut ML. 2005. Changes in immunomodulatory properties of *Echinacea* spp. root infusions and tinctures stored at 4°C for four days. Clinica Chimica Acta. 355:67-82.

Wu L, Bae J, Kraus, GA, Wurtele ES. 2004. Diacetylenic isobutylamides of *Echinacea*: Synthesis and natural distribution." *Phytochemistry*. *65*, 2477.

I. Oilseed Crops (L. Marek, I. Larsen, B. Bingaman)

Acquisitions/collection maintenance:
In 2005, we received 115 new oil seed accessions.

Helianthus:
Nine cultivated *H. annuus* accessions under CSR were received from NCGRP. Two *H. annuus* disease resistant populations derived from recurrent selection of bulked wild accessions were released by Dr. Charles Block, NCRPIS, in January 2005 and incorporated into the collection for distribution. Three *H. niveus* ssp *tephrodes* accessions were collected in California in December 2004 by Dr. Laura Marek and BLM cooperator Mr. Chris Knauf. Ten accessions of *H. niveus* ssp *tephrodes* and *H. niveus* ssp *canescens* were collected in California and Arizona in late February and early March 2005 by Dr. Marek, USDA Sunflower researchers Dr. Gerald Seiler and Dr. Thomas Gulya, and BLM cooperator Mr. Chris Knauf. Nine accessions of *H. cusickii* were collected in Washington and Oregon in early June 2005 by Dr. Marek and Dr. Joe-Ann McCoy. Six accessions of *H. deserticola* and nine accessions of *H. cusickii* were collected in California and Nevada in mid-June by Dr. Gulya. Forty-five accessions of *H. pumilus* and one each of *H. annuus* and *H. nuttallii* ssp *nuttallii* were collected in Colorado and Wyoming in August by Dr. Marek, Dr. Seiler, and Dr. Gulya. Three additional *H. pumilus* accessions from Colorado were collected by two cooperators and donated in August. Seven *H. deserticola* accessions were collected in Nevada in July by a cooperating BLM scientist and donated in August. One *H. carnosus* accession, collected by botanists from the Bok Historic Sanctuary in Florida, was donated in late November 2005. The new sunflower accessions include twelve taxa, five of which previously had no accessions available for distribution. An increased interest by the sunflower research community in wild *Helianthus* germplasm has resulted in the CGC approved goal of acquiring accessions representative of the complete geographic distribution of all 66 Helianthus taxa.
Miscellaneous asters:
One new *Heliopsis helianthoides* accession was received from the NRCS. The accession is composed of a bulk collection from 48 locations of remnant prairie in the southern third of the counties in Iowa.

Brassicaceae:
Seven *Brassica napus* accessions originally received by the NPGS in 1985 and 1986 from Svalof Weibull AB, Sweden came off PVP protection in 2005 and were received from NCGRP. One *Brassica napus* still under CSR protection was received from the University of Idaho.

Cuphea:
One accession of *Cuphea carthagenensis* was collected in North Carolina by Dr. Joe-Ann McCoy during the fall of 2005.

Maintenance:
General statistics about availability and management of the collections are presented in Tables 1 and 2 in the appendix.

As part of a major effort to ensure that the active oil seed collection contains only viable accessions and accessions for which we can distribute seeds, one hundred eighty four wild *Helianthus* accessions were prepared for inactivation in late 2005. These accessions were inactivated because none to fewer than ten seeds remain and one to several germination attempts failed. The inactivated accessions were from the wild collection and 88% had original seeds collected prior to 1980. I anticipate that the 2006 and 2007 increase attempts will result in inactivation of most of the remaining non-viable *Helianthus* accessions. Similar collection analyses will be initiated for the Brassicaceae and Cuphea.

Helianthus:
The cultivated *H. annuus* accessions are 87% available. When CSR restricted accessions are not included, cultivated *Helianthus* accessions are 94% available. We are managing our increases to maintain that level of availability and to ensure that the core collection accessions are always available. Cultivated *H. annuus* accessions requiring long seasons or short days to flower are increased in the greenhouse as space allows (three to four accessions per season; four in the winter of 2005-2006). Research cooperators are strongly interested in wild *Helianthus* germplasm as potential sources of disease resistance, herbicide tolerance and other valuable traits. As a result, we have substantially increased our efforts to make wild annual and perennial accessions available at the same level as the cultivated accessions. Wild annual *Helianthus* accessions are 84% available and wild perennial accessions are 40% available. We caged and harvested seed from 22 wild annual *Helianthus* accessions and 127 wild perennial accessions in 2005. Thirty-three of the caged wild perennial accessions were populations established in the field from 2000 through 2003. Thirty-four of the wild perennial accessions were established in the field in 2004 and caged for the first time in 2005. Sixty-four perennial accessions were established in the field in 2005 and 58 of these were caged for seed production in 2005. The 64 new accessions represent 58% of the wild perennials that we attempted to regenerate in 2005. This is an increase in the success rate of germination efforts in 2004 when 46% of the attempted wild accessions established in the field.
We are working with the NPGS Parlier site (Dr. Maria Jenderek, curator) to regenerate accessions that require a longer growing season than is available in Ames and for a portion of our cold sensitive wild *Helianthus* accessions. Not all cold sensitive accessions can be successfully grown in Parlier because the location averages 30 occurrences annually of temperatures at or below -3.0ºC. Because of the difficulties associated with germinating wild *Helianthus* species, to facilitate our interactions with Parlier in 2005, we adopted the protocol of germinating seeds for Parlier increases in Ames and shipping live seedlings to California. The Parlier crew shipped harvested sunflower material to Ames for threshing and cleaning. It is beneficial for us to observe intact seed heads in order to collect evaluation information prior to processing. Our new management strategy was very successful in 2005; 27 of 29 accessions sent to Parlier in 2005 were harvested, a significant increase over the 12 of 29 accessions harvested in 2004. We will use the 2005 protocol for future increases at Parlier, sending seedlings to California by early April each year. The NCRPIS has purchased 40 sunflower cages to enable Parlier to handle up to 40 accessions per year.

Brassicaceae:
Brassicaceae accessions are 80% available; our goal is to have 90% or more of these accessions available within the next ten years. We harvested 156 accessions in 2005; 51 accessions did not flower in the field. Up to 20 plants from each of 39 of these 51 accessions were transplanted to over-

winter in the greenhouse. Transplanting was a priority for accessions which had no remaining seed and/or those for which we did not have useful information about winter survival.

Due to greenhouse space constraints, if an accession had more than 20 plants, a portion of the population was left in the field. There may be interest in some of the wild *Brassicaceae* as ornamentals; evaluation of winter survival characteristics would provide valuable information. Several of the transplanted accessions began flowering during the late fall; additional accessions are expected to bloom as the days lengthen. Many of the wild *Brassicaceae* are of Mediterranean origin and could be expected to bloom during cool, moist, short-day winter weather. We were successful with winter greenhouse caging of field to greenhouse transplanted *Erysimum* accessions during 2004-2005 and will continue to use this management tool as needed. Six of the *Erysimum* accessions flowered during the 2004-2005 winter season and increases were obtained after caging and introducing pollinators.

<u>*Linum*</u>:
Fifteen of seventeen *Linum usitatissimum* accessions planted in 2005 were successfully harvested. The two failed accessions will be replanted in 2006. Fourteen of the 2005 planted accessions had not been increased previously and represent newly available genetic material for the scientific community. Nine of these new accessions were collected in 2004 in Tajikistan and five accessions were received from China in 1991.

No wild flax accessions were over-wintered from 2004. Thirty wild flax accessions were planted in the field in 2005. Seven of these were harvested, two of which produced enough seed to make the accessions available. Up to 20 plants of accessions for which we could not be certain would over-winter and/or which had no remaining seed were moved to the greenhouse before hard freezes in the fall. Due to greenhouse space constraints, if a population had more than 20 plants a portion of the population was left in the field to test for winter survival. A total of 23 accessions were transplanted to the greenhouse; nine of these accessions were also represented by field populations. An additional six accessions were left in the field to over-winter giving a total of 15 over-wintering populations.

Cultivated flax accessions are 99% available. Wild Linum accessions are currently 40% available; our goal is to make more than 90% of these accessions available within the next ten years. We attempted to increase 65% of the unavailable wild *Linum* accessions in 2005; 52% of these did not germinate. Of the 30 accessions for which field populations were established, 2 accessions yielded enough seeds to make them available. We look for successful harvest of the remaining wild *Linum* accessions during 2006.

<u>*Cuphea*</u>:
Seeds are available for 89% of the accessions of seven species (*C. calophylla, C. carthegenensis, C. lanceolata, C. lutea, C. tolucana, C. viscosissima, C. wrightii*) being used by members of the National Cuphea Consortium for the development of domesticated lines of cuphea as a source of mid-chain fatty acids. Seeds were harvested from all 27 accessions established in the field in 2005. Fifty-four percent of the cuphea accessions of species of primary interest for horticultural/floricultural uses are available. Selections from five of 55 accessions sent to the University of Georgia in 2005 are entered in the prestigious Allan Armitage Trial Gardens for evaluation as ornamentals. Due to steady interest in cuphea by horticultural groups, we are working to make a larger percentage of non-consortium targeted accessions available.

Distributions:
General statistics about distributions of the collections are presented in Tables 3A and 3B.

Helianthus:
The increase in both cultivated and wild helianthus distributions in 2005 was largely due to a request from Embrapa-Cenargen, Brazil (1175 accessions). We distributed helianthus accessions in 2005 to cooperators at the USDA Sunflower Research Group in Fargo for evaluation of resistance to sunflower moth, banded sunflower moth, red sunflower seed weevil, resistance to multiple races of rust and downy mildew, and for oil evaluations. Accessions have been distributed to a wide range of research projects including on-going work at Indiana University on control of flowering, genetic research at Vanderbilt University and drought tolerance at the University of Agriculture in Faisalabad, Pakistan. Other accessions were requested for use in investigations of polyploid evolution, biomass production and resistance to *Orobanche cumana*. In addition, NRPIS continues to supply germplasm to be used for the *Helianthus* portion of the National Science Foundation funded *Compositae* Genome sequencing project.

Brassicaceae:
Demand for Brassicaceae accessions continued to be strong in 2005 with 1489 accessions sent out for general breeding research and 1155 accessions distributed for disease screenings. Portions of the *Brassicaceae* collection were distributed for phytoremediation research, tissue culture research and biofumigant and green manure studies. In addition, the *Camelina* collection was distributed for research on oil content and biolubricant development.

Linum:
We distributed *Linum* accessions to several breeding programs and to a new research program working to develop biomarkers to characterize flax domestication. All accessions of Egyptian origin were repatriated to Egypt for incorporation into a new genebank.

Cuphea:
Cuphea accessions were distributed to breeding and evaluation programs participating in the National Cuphea Consortium, for molecular systematic research at St Louis University and for ornamental evaluations at several locations.

Research Activities:
General observation statistics and images recorded for the collections are presented in Table 4.

Helianthus:

Germination experiments: Increases have not been attempted for most wild perennial accessions and 65% of the original seed has been stored for 19 years or longer. One of our biggest issues in the germination of the wild sunflowers is pathogen contamination of old original seed. We have incorporated a five minute soak in 3% hydrogen peroxide followed by two minutes of rinsing in cool, running tap water to help control the pathogens prior to our standard pre-germination cold incubation period followed by transfer of the accessions to germination temperatures. We continue to modify our protocols to find optimal germination conditions for the different species.

Molecular profiling: I have started preliminary work to establish molecular profiles of the *H. niveus* species group. Tissue samples were captured on Whatman FTA™ cards during collection trips. DNA can be extracted from the cards for use in intra- and inter-population variation studies. As protocols are established, we will also examine molecular profiles of our *H. tuberosus* collection to determine the genetic integrity of accessions that have been maintained long term in a perennial nursery.

Herbicide tolerance: We participated in an herbicide tolerance screening project with Dr. Mike Owen, Agronomy, ISU for Dr. Mark Dahmer, BASF. A select group of wild *H. annuus and H. petiolaris* species collected in Canada were screened with the herbicide imazamox (Beyond). Seedlings grown at NCRPIS were transferred to an ISU greenhouse, where Dr. Owen's group conducted the herbicide screen. During the same time period, the BASF laboratory in North Carolina ran an identical experiment. Both experiments gave similar results which allowed BASF to satisfy Canadian government requirements to allow the marketing of Clearfield sunflowers.

Brassicaceae:

Germplasm for phytoremedition: The field population established in 2004 of a Ni accumulating *Alyssum murale* accession donated by Dr. Rufus Cheney over-wintered successfully. It was left in the field for the 2005-2006 winter to monitor survival potential in Iowa.

Brassica napus genetic study: Ten of the 40 accessions of *Brassica napus* grown in 2004 to provide seed for a genetic study by graduate student Mark Cruz did not flower and were left in the field to overwinter. Only a few scattered plants showed re-growth in the spring. These 10 accessions were germinated again in 2005 and received a longer vernalization treatment. Summer flowering was not uniform; only a few plants bloomed of several accessions. Seed was harvested from eight of the accessions, although in low amounts for all but two accessions. A few plants of representative *B. napus* accessions were transplanted to pots, cut back and moved to the greenhouse for the winter of 2005-2006. If these plants bolt, we plan to start these 10 accessions again early in early fall 2006 and grow them in the cool, moist "Mediterranean winter" conditions of our "bubble" greenhouse to provide an extended vernalization treatment prior to field growth.

Eruca collection from Italy: In 2004, NPGS received 59 *Eruca sativa* accessions collected primarily in Italy as market samples. The scientific community working with *Eruca* is relatively small but strongly interested in new material that might offer disease and/or insect resistance. We attempted to increase all of the new *Eruca* accessions as well as a selection of geographically diverse *Eruca* accessions already in the collection. Fifty-seven of the new accessions germinated, were established in the field and harvested. To preserve DNA for future molecular assessment of the collection, four random plants were sampled from each accession using Whatman FTA cards.

Cuphea:

Germination experiments: Based on results from 2004 experiments, our germination protocol was modified to eliminate seed coat excision after imbibition but prior to germination. Using the same species and seed lots as we used for our 2004 experiments, we continued to test different combinations of treatments in 2005 for which we observed a positive effect on germination. For *C. wrightii* and *C. carthagenensis*, if seed had been frozen, including a pre-imbibition heat treatment improved germination. The heat treatment did not have a positive effect on *C. lanceolata* or *C. viscosissima* germination. We are continuing our experiments to find optimal germination conditions, especially for *C. viscosissima* which seems to be losing viability more quickly than the three other species used in these experiments.

Harvest equipment testing: The Cuphea research group continues efforts to domesticate PSR23 and enable full agronomic production. In collaboration with Dr. Win Phippen, we grew a half acre of the Western Illinois University PSR23 line to provide raw material for testing modifications to harvest equipment. Dr. Phippen came from Illinois to plant the cuphea; NCRPIS managed the plot from that point, including weeding and spraying desiccant prior to the harvest. The cuphea grew very well in Ames during a summer with somewhat below average precipitation.

Collection trips:
I participated in three collection trips for wild *Helianthus* germplasm in 2005. In February/March, I met Dr. Gerald Seiler, Botanist, and Dr. Thomas Gulya, Plant Pathologist, from the USDA Sunflower Research Group, Fargo in southern California. We spent 5 days collecting wild *Helianthus* germplasm from the Imperial Sand Dunes in southeast California, and from the Yuma area and the Cabeza Prieta Wildlife Refuge in southwest Arizona. We collected accessions of *H. niveus* ssp *tephrodes* (3), *H. niveus* (1), *H. niveus* ssp *canescens* (3) and *H. annuus* (1). In June, I collected 9 accessions of *H. cusickii* (2 populations from south-central Washington and 7 populations from north-central and east-central Oregon). Dr. Joe-Ann McCoy, Medicinals Curator, NCRPIS, accompanied me during the Oregon portion of the trip. In August, I met Dr. Seiler and Dr. Gulya in Denver, Colorado. We spent 13 days collecting 45 accessions of *H. pumilus,* one accession of *H. nuttallii* ssp *nuttallii* and one accession of *H. annuus* throughout the foothills and adjacent plains in Colorado and Wyoming. Dr. Mark Dahmer, National Sunflower Association Board member, participated for three days. Our collection sites ranged from the southern Sangre de Cristo Mountains southwest of Westcliffe, Colorado to the edge of the Big Horn National Forest just west of Dayton in north central Wyoming.

Professional activities:
Training:
During August and September 2005 I spent time with Dr. David James, USDA-ARS Soil Tilth Laboratory, Ames, IA learning how to use the ArcGIS 9 software package. Mapping technologies are essential for collection distribution analyses supporting collection trip planning and reporting.

Publications and Presentations:

Helianthus: I presented a talk entitled "Current Status of the Wild *Helianthus* Seed Collection" at the National Sunflower Association's 27th Annual Research Forum in Fargo in January 2005. I presented the biannual *Helianthus* Germplasm Status Report to the Sunflower CGC committee, also in Fargo in January.

Brassicaceae: I prepared the annual *Brassicaceae* Germplasm Status Report for the Brassica CGC committee for presentation during the ASHS Meeting in Las Vegas, NV in July.

New Crops: I presented an overview of the Cuphea collection and discussed ongoing germination experiments at the Cuphea Winter Technical Meeting in Athens, GA, February 1-2. I presented a Crop Status Report including data for the *Cuphea*, *Vernonia* and *Euphorbia* collections to the New Crops CGC meeting at the ASA/CSSA/SSA annual meeting in Salt Lake City, UT in November.

NPGS Curators' Workshop: I was co-organizer for the Evaluation Data segment of the 1st Biennial NPGS Curators' Workshop and presented a talk entitled "Molecular Marker Evaluation Data".

Service Activities:
NCRPIS:
I am a member of the Safety Committee (inactive in 2005).

Agronomy Departmental activities:
I continue to coordinate the monthly Agronomy Department Professional and Scientific staff meetings. Following an agenda pattern established in 2004-2005, the group continues to have informal monthly presentations by P&S staff from the different research/teaching/service groups within the department describing their area activities after the main agenda discussion usually led by the department chair. NCRPIS P&S staff presented in fall of 2004.
PGOC:
I attended the PGOC annual meeting in Pullman, WA in June. I am a member of the recently re-activated *In situ* Conservation Subcommittee within the PGOC which held its first formal meeting since re-activation at the Pullman meeting.

J. Vegetables (K. Reitsma, L. Clark)

Collections curated by the Vegetable Project include *Cichorium* (NC7-chicory), *Cucumis sativus* (NC7-cucumis.cucs), *Cucumis melo* (NC7-cucumis.melo), *Cucumis* species (NC7-cucumis.wilds), *Cucurbita pepo* (NC7-cucurbita), *Cucumella aspera and Oreosyce africana* (NC7-cucurbits.misc), *Daucus* (NC7-daucus), *Ocimum* (NC7-ocimum), and *Pastinaca* (NC7-parsnips). Statistics for accession numbers and availability for each site crop are found in Table 1.

Acquisition:
Seven new accessions were received and are listed by site-crop in Table 1. The new accessions include three *Cucumis melo* and two *C. sativus* accessions received from NCGRP; a new Ames number was assigned to the *C. myriocarpus* portion separated from an accession of *C. anguria*; and a new Ames number was assigned to the annual portion of a biennial petaloid Ames-numbered *Daucus carota* accession.

Maintenance:
Table 2 contains 2005 data for regenerations attempted and accessions harvested.

Cucurbit regenerations focused on accessions having low seed quantities on distribution lots. Many of the *Cucumis melo* grown in 2005 were accessions which produced an insufficient quantity of seeds during the 2004 regenerations. In May 2005, the *C. melo* accessions were direct seeded into peat pots in the campus greenhouse where individual flats could be isolated from each other by distance to reduce the possibility of spreading *Acidovorax avenae* subsp. *citrulli*(bacterial fruit blotch), a seed-borne disease primarily spread when bacteria are splashed via watering. To further reduce the chance of spreading the disease, a new bottom-watering system was established where each flat containing the peat pots was placed in a second flat which served as a reservoir for water. The reservoirs were carefully filled with water so as not to allow the foliage to become wet. Seedlings were observed daily for symptoms of bacterial fruit blotch. No bacterial fruit blotch was found on the melon seedlings in the greenhouse or on plants after transplanting to field cages; therefore, no hydrochloric acid treatment of seeds was necessary at harvest.

Twenty-two 5' x 15' x 40' *Cucurbita* field cages were used in the summer of 2005. We continue to monitor the effectiveness of the cage program in reducing the incidence of and/or delaying the transmission of squash mosaic virus. (For additional information on current NCRPIS research activities on bacterial fruit blotch and squash mosaic virus, please refer to the Plant Pathology section of this annual report.)

Daucus regeneration efforts included new accessions and older accessions with low seed quantities or viabilities. In addition to the regenerations in Ames, we received seed increases from R. Maxwell, Seminis Vegetable Seeds, Idaho (3 accessions), and P. Simon, University of Wisconsin, Madison, WI (17 accessions).

As NCRPIS accessions are regenerated, seed samples are sent to NCGRP for back-up. Seven of the nine vegetable collections have 79% or more of their accessions backed up at NCGRP (Table 2).

In 2005, 940 germination tests (Table 2) were performed, and included seed increases from the 2004 regenerations and 5-year viability testing of *Cucurbita* and *Daucus* accessions.

Distribution:
Packet and accession distributions for the vegetable collections are summarized in Table 3A and 3B. In 2005, 7291 packets (items) were distributed for 241 domestic and foreign orders. This represented 3551 vegetable accessions. Distribution history of the vegetable crops for the last five years can be found in the following table.

Crop	Calendar Year	No. of Orders	No. of Recipients	No. of Items Distributed	No. of Accessions Distributed
Cichorium	2001	6	6	288	175
	2002	8	8	261	134
	2003	8	7	195	144
	2004	5	4	45	43
	2005	9	9	257	118
Cucumis	2001	59	49	1229	933
	2002	58	46	2658	1773
	2003	46	36	1901	1391
	2004	73	64	1393	1105
	2005	101	89	4768	2142
Cucurbita	2001	22	20	288	156
	2002	20	17	165	132
	2003	11	11	170	150
	2004	38	35	702	490
	2005	51	49	1568	829
Cucurbits -Misc.	2001	2	2	2	1
	2002	0	0	0	0
	2003	1	1	1	1
	2004	1	1	2	2
	2005	2	2	2	1
Daucus	2001	13	12	235	211
	2002	11	11	75	67
	2003	13	12	426	294
	2004	21	21	596	378
	2005	23	23	491	375
Ocimum	2001	4	4	96	79
	2002	7	7	18	16
	2003	8	8	52	42
	2004	22	22	235	84
	2005	23	21	204	85
Pastinaca	2001	0	0	0	0
	2002	2	2	9	8
	2003	1	1	1	1
	2004	3	3	5	4
	2005	1	1	1	1
Total		673	604	18336	11365

Characterization and Taxonomy:
Digital images, along with basic notes for taxonomic identification and accession characterization, are recorded during regeneration (Table 4). Data for approximately 17 descriptors, primarily fruit descriptors, are recorded at harvest for *Cucumis* and *Cucurbita* accessions. Plant habit, flowering dates, and life-cycle notes are recorded for *Daucus* and *Pastinaca*. Some fruit images of *Cucumis* have been loaded to GRIN, and more fruit images of C*ucumis* and *Cucurbita* will be loaded to GRIN early in 2006.

With the assistance of Dr. Mark Widrlechner (Horticulturist), taxonomic identities are reviewed and confirmed as each accession is regenerated. The 2005 re-identifications included: 2 *C. anguria* to *C. anguria* var. *anguria*, 2 *C. anguria* to *C. anguria* var. *longaculetus*, 1 *Cucumis melo* to *Cucumis sp.*, 1 *C. sativus* to *C. sativus* var. *hardwickii*, 2 *C. sp.* to *C. ficifolius*, 1 *C. zeyheri* to *C. myriocarpus*. We are attempting to describe and identify one Cucumis sp. accession which may be a previously un-described species.

Evaluation/Utilization:
Dr. Charles Block (Pathologist) continued to screen all *Cucurbita* and *Cucumis* seedlings grown for regeneration for the presence of squash mosaic virus, using ELISA protocols, before seedlings are transplanted to the field. He also visually inspected all cucurbit field plantings for disease during the 2005 growing season. Seed-borne diseases are of specific interest, with bacterial fruit blotch in *Cucumis melo* being of greatest concern since phytosanitary issues have prevented the distribution of *Cucumis* germplasm to some countries. This year we encountered angular leaf spot (*Pseudomonas syringae* pv. *lachyrmans*) in some of the *Cucumis* cages – a disease which we have not seen in this area for at least 15 years.

In collaboration with Larry Lockhart (Program Manager) and Brian Buzzell (Farm Equipment Mechanic), the Pathology and Vegetable Projects designed a barrier system to reduce the transmission of *Acidovorax avenae* subsp. *citrulli* (bacterial fruit blotch), a disease primarily spread by bacteria being splashed during watering. The barrier system allows for the isolation of individual flats of *Cucumis melo* in a small space in the greenhouse and, when combined with a bottom-watering reservoir system, significantly reduces transmission of bacterial fruit blotch.

The Pollinator Program and the Vegetable Program also continue to collaborate on pollinator tests. One test involves evaluating whether alfalfa leaf cutter bees may be a useful pollinator for *Cucumis melo*, *Cucumis sativus*, *Cucurbita pepo*, *Daucus*, and *Ocimum* in field and greenhouse regenerations. Collaboration continues on developing a year-round cage and pollinator program for regenerating *Cucumis* and *Cucurbita* in the greenhouse. For more information on these tests, please refer to the Entomology section of the annual report.

Plans for 2006:
Regenerations: Thirty accessions of *Daucus* were started in the greenhouse in October 2005 for the summer 2006 field cages. In January 2006, all unavailable *Cichorium* will be direct seeded into flats in the greenhouse for regeneration in field cages in 2006. Regeneration of hard-to-handle and wild *Cucumis* species will continue in the greenhouse as time, space, and labor permit. We will continue to increase *Cucumis* and *Cucurbit* accessions where distribution quantities and viabilities have fallen below critical values as set on GRIN. The *Cucumis melo* will be started in the greenhouse using a barrier system to isolate individual accessions and a reservoir watering system to prevent the transmission of *Acidovorax avenae*.

Germinations: Viability tests will be performed on the 2005 cucurbit regeneration seed lots in April 2006 and on the 2005 *Daucus* regeneration seed lots in June 2006.

Characterization: A 2-year observation planting of all available *Cichorium* accessions will be direct seeded in the spring of 2006 so that the collection may be characterized and accession taxonomy verified. Resulting data will be loaded into GRIN. We will also continue to record characterization data as regenerations occur on other vegetable accessions.

Evaluation: The Pollinator Program and the Vegetable Program will continue to collaborate on pollinator tests evaluating whether alfalfa leaf cutter bees may be a useful pollinator for vegetable crops maintained at the NCRPIS in field and greenhouse regenerations. Collaboration continues on developing a year-round cage and pollinator program for regenerating *Cucumis* and *Cucurbita* in the greenhouse. For more information on these tests, please refer to the Entomology section of the annual report. In 2006, we will continue to investigate improvement of insect pollinator effectiveness in the large *Cucurbita* cages by introducing two honey bee nucleus hives (one each at the north and south ends of the cages), raising the nucleus hives above the canopy of foliage, and trying *Bombus* (bumblebees) as pollinators.

The Pathology Project and the Vegetable Project will continue to monitor the effectiveness of the cage program in reducing the incidence of and/or delaying the transmission of squash mosaic virus and other insect vectored diseases in the cucurbits.

Another possible evaluation project for the future will be to compare the OP melon seed lots transferred to Ames with seed samples backed up at NCGRP by the SRPIS. Many of the older seed lots were backed up prior to 1970 and may have been regenerated by open pollination less often than the seed samples sent to Ames. Unfortunately, many of these backup samples are low quality and low quantity and will have to be regenerated before we can do the comparisons.

Publications:
The following article was published in 2005:
Block, C. C. and K. R. Reitsma. Powdery Mildew Resistance in the U.S. National Plant Germplasm System Cucumber Collection. HortScience 40(2) 416-420. 2005.

K. Research Leader Activities (C. Gardner)

Primary duties include the coordination of the ARS and NC7 components of the North Central Regional Plant Introduction Station, including the Plant Introduction Research and GEM CRIS Projects. In 2004-2005, I served as mentor for Borlaug Fellow Ana Gulbani from the Republic of Georgia. Ana spent 18 months with us learning about germplasm conservation principles, methods and practices. She returned to Georgia in December, 2005, full of knowledge and enthusiasm, but to a largely unfunded Georgian Germplasm Institute. We hope the Institute's funding becomes more stable.

Current research activities/interests include:
1) Development of best pollination practices guidelines to preserve genetic profile during regeneration practices.
2) Oversight of Plant Breeding Ph.D. student Von Mark Cruz' Brassica genetics studies.
3) Co-advises Sustainable Agriculture M.S. student Lindsay Werth's Southwestern Maize racial characterization project.
4) Research interests in the development of use of alfalfa leafcutter bees for NCRPIS crop regenerations and development of medicinal crop plants collections.
5) Improving the software tools available to public and internal users of GRIN
6) Serves on ASA's Biosecurity Committee and secretary for AAIC, Am. Assoc. of Industrial Crops
8) Coordinating collection of site needs for the ARS's NPGS greenhouse initiative
9) Co-chair for the 9th International Plant – Pollinator Relations Symposium in Ames, IA in 2007.

C. Gardner participated in data capture and harvest of graduate student Lindsay Werth's thesis trials at the New Mexico Agricultural Experiment Station in October, 2005. She traveled to Murcia, Spain in September to participate in the AAIC (American Association of Industrial Crops) International Meeting and presented a poster on the plant genetic resources held at the NCRPIS and chaired a session on alternative crop research. She also made invited presentations on the NCRPIS programs to students at the Ames, IA High School.

Peer-Reviewed 2005 Publications:

Crane, J., D. Kovach, C. Gardner and C. Walters. Accepted 2005. Triacylglycerol phase and 'intermediate' seed storage physiology: a study of Cuphea carthagenensis. Planta.

Year 2005 Table 1 NCRPIS Accessions (Acc), Acquired, Available

CURATOR	GENUS CROP	Number Accs	Number Accs Acquired	Percent Acquired	Number Available	Percent Available	Percent Avail Last Year
Brenner	NC7-amaranth	3330	1	0	3136	94	93
	NC7-celosia	54	0	0	23	43	43
	NC7-echinochloa	302	0	0	236	78	72
	NC7-grasses	119	0	0	74	62	12
	NC7-legumes	227	0	0	105	46	48
	NC7-melilotus	952	0	0	687	72	71
	NC7-panicum	948	0	0	871	92	89
	NC7-perilla	22	0	0	22	100	100
	NC7-quinoa	234	0	0	189	81	82
	NC7-setaria	1006	0	0	892	89	89
	NC7-spinach	401	0	0	377	94	92
	NC7-umbels	1069	15	1	524	49	43
	Total:	8664	16	0	7136	82	80
Marek	NC7-asters	324	1	0	91	28	28
	NC7-brassica	2001	8	0	1717	86	83
	NC7-crucifers	1197	0	0	776	65	61
	NC7-crucifers.pvp	1	0	0	0	0	0
	NC7-cuphea	650	0	0	465	72	73
	NC7-euphorbia	219	0	0	46	21	21
	NC7-flax	2818	0	0	2798	99	99
	NC7-flax.wilds	165	0	0	64	39	39
	NC7-sun.cults	1690	9	1	1519	90	90
	NC7-sun.wilds	24	0	0	12	50	50
	NC7-sun.wilds.ann	1340	19	1	1132	84	77
	NC7-sun.wilds.per	679	78	13	231	34	20
	Total:	11108	115	1	8851	80	77
McCoy	NC7-medicinals	305	59	19	152	50	38
	Total:	305	59	19	152	50	38
Millard	NC7-corn.kin	34	0	0	5	15	21
	NC7-maize	18370	75	0	12145	66	65
	Total:	18404	75	0	12150	66	64
Reitsma	NC7-chicory	250	0	0	195	78	78
	NC7-cucumis.cucs	1351	2	0	1280	95	95
	NC7-cucumis.melo	3106	3	0	2378	77	75
	NC7-cucumis.wilds	332	1	0	142	43	39
	NC7-cucurbita	986	0	0	807	82	82
	NC7-cucurbits.misc	2	0	0	2	100	100
	NC7-daucus	1087	1	0	844	78	68
	NC7-ocimum	96	0	0	85	89	89
	NC7-parsnips	71	0	0	51	72	51
	Total:	7281	7	0	5784	79	75
Widrlechner	NC7-mints	128	0	0	72	56	59
	NC7-ornamentals	1980	29	1	852	43	43
	Total:	2108	29	1	924	44	44
NCRPIS Total:		**47870**	**301**	**1**	**34997**	**73**	**71**

Year 2005 Table 2 **NCRPIS Accessions (Accs) Germinated, Regenerated, Made Available, Backed Up**

CURATOR	GENUS_CROP	Number Accessions (Accs)	Number Accs Germed	Percent Accs Germed	Number Attempted Regen	Number Harvested Regen	Number Perm Perennial	Number Perennial Harvested (Vegetative)	Number Accs Made Available	Number Accs Growing	Number Accs Backed Up for YR	Total Number Accs Backed Up	Percent Accs Backed Up
Brenner	NC7-amaranth	3330	567	17	141	62	0	0	83	0	68	3131	94
	NC7-celosia	54	0	0	5	4	0	0	0	0	0	23	43
	NC7-echinochloa	302	217	72	68	12	0	0	33	0	27	224	74
	NC7-grasses	119	86	72	14	12	0	0	0	0	36	74	62
	NC7-legumes	227	0	0	1	1	0	0	1	0	0	168	74
	NC7-melilotus	952	24	3	25	5	0	0	23	0	17	773	81
	NC7-panicum	948	0	0	24	18	0	0	0	0	0	888	94
	NC7-perilla	22	0	0	0	0	0	0	0	0	0	22	100
	NC7-quinoa	234	0	0	1	13	0	0	0	0	0	201	86
	NC7-setaria	1006	247	25	1	1	0	0	0	0	0	944	94
	NC7-spinach	401	12	3	0	14	0	0	10	0	3	374	93
	NC7-umbels	1069	74	7	72	25	0	0	81	30	75	541	51
	Total:	8664	1227	14	352	167	0	0	231	30	226	7363	85
Marek	NC7-asters	324	0	0	0	0	0	0	1	0	1	73	23
	NC7-brassica	2001	251	13	57	37	0	0	52	4	36	1944	97
	NC7-crucifers	1197	64	5	204	118	0	0	61	31	55	849	71
	NC7-crucifers.pvp	1	0	0	0	0	0	0	0	0	0	1	100
	NC7-cuphea	650	0	0	0	0	0	0	7	0	3	563	87
	NC7-euphorbia	219	2	1	0	0	0	0	0	0	0	53	24
	NC7-flax	2818	0	0	0	12	0	0	0	0	0	2816	100
	NC7-flax.wilds	165	12	7	65	7	0	0	13	28	6	60	36
	NC7-sun.cults	1690	51	3	0	0	0	0	43	0	24	1600	95
	NC7-sun.wilds	24	0	0	0	0	0	0	0	0	0	13	54
	NC7-sun.wilds.ann	1340	23	2	0	22	0	0	34	0	31	1144	85
	NC7-sun.wilds.per	679	91	13	0	0	1	0	107	0	87	251	37
	Total:	11108	494	4	326	196	1	0	318	63	243	9367	84
McCoy	NC7-medicinals	305	73	24	86	50	0	1	42	153	54	152	50
	Total:	305	73	24	86	50	0	1	42	153	54	152	50
Millard	NC7-corn.kin	34	0	0	1	0	0	0	1	0	1	8	24
	NC7-maize	18370	1332	7	294	301	0	0	436	7	76	13601	74
	Total:	18404	1332	7	295	301	0	0	437	7	77	13609	74
Reitsma	NC7-chicory	250	0	0	0	0	0	0	0	0	0	199	80
	NC7-cucumis.cucs	1351	272	20	60	51	0	0	39	0	29	1271	94
	NC7-cucumis.melo	3106	129	4	50	50	0	0	127	0	70	2484	80
	NC7-cucumis.wilds	332	19	6	1	2	0	0	15	0	17	149	45
	NC7-cucurbita	986	128	13	37	26	0	0	21	0	18	783	79
	NC7-cucurbits.misc	2	0	0	0	0	0	0	0	0	0	2	100
	NC7-daucus	1087	373	34	30	59	0	0	114	0	52	859	79
	NC7-ocimum	96	4	4	14	6	0	0	0	0	0	87	91
	NC7-parsnips	71	15	21	0	1	0	0	15	0	15	47	66
	Total:	7281	940	13	192	195	0	0	331	0	201	5881	81
Widrlechner	NC7-mints	128	4	3	33	13	0	0	0	0	0	76	59
	NC7-ornamentals	1980	54	3	147	67	72	69	53	7	38	630	32
	Total:	2108	58	3	180	80	72	69	53	7	38	706	33
NCRPIS Total:		**47870**	**4124**	**9**	**1431**	**989**	**73**	**70**	**1412**	**260**	**839**	**37078**	**77**

Year
2005 **Table 3A** **External NCRPIS Distributions**

			External Domestic Distributions				Foreign Distributions				External Domestic & Foreign Distributions				
CURATOR	**GENUS CROP**	**# Accs**	**# Accs**	**# Orders**	**# Recipients**	**# Items**	**# Accs**	**# Orders**	**# Recipients**	**# Items**	**# Accs**	**# Orders**	**# Recipients**	**# Items**	**# Distinct Accs for NC7 Orders**
Brenner	NC7-amaranth	3330	186	44	39	382	114	15	14	158	229	59	53	540	711
	NC7-celosia	54	6	4	4	6	1	1	1	1	6	5	5	7	9
	NC7-echinochloa	302	56	3	3	56	3	2	2	3	57	5	5	59	268
	NC7-grasses	119	3	1	1	3	0	0	0	0	3	1	1	3	89
	NC7-legumes	227	22	5	4	26	2	2	2	2	24	7	6	28	1
	NC7-melilotus	952	45	9	7	49	30	7	7	34	73	16	14	83	52
	NC7-panicum	948	8	5	5	9	65	6	6	71	68	11	11	80	73
	NC7-perilla	22	22	8	8	52	15	3	3	22	22	11	11	74	1
	NC7-quinoa	234	72	24	22	118	113	8	8	184	138	32	30	302	8
	NC7-setaria	1006	12	2	2	13	123	8	7	137	126	10	9	150	338
	NC7-spinach	401	370	10	10	392	7	2	2	7	370	12	12	399	103
	NC7-umbels	1069	211	22	21	229	9	5	4	10	215	27	25	239	273
	Total:	8664	1013	137	126	1335	482	59	56	629	1331	196	182	1964	1926
Marek	NC7-asters	324	68	7	7	70	3	2	2	3	70	9	9	73	1
	NC7-brassica	2001	643	34	33	725	808	22	21	963	1286	56	54	1688	221
	NC7-crucifers	1197	467	37	33	809	275	15	15	677	497	52	48	1486	350
	NC7-crucifers.pvp	1	0	0	0	0	0	0	0	0	0	0	0	0	0
	NC7-cuphea	650	135	18	11	171	209	2	2	280	277	20	13	451	41
	NC7-euphorbia	219	1	1	1	1	0	0	0	0	1	1	1	1	7
	NC7-flax	2818	1365	6	6	1373	168	7	7	173	1441	13	13	1546	14
	NC7-flax.wilds	165	30	5	5	32	2	1	1	2	30	6	6	34	87
	NC7-sun.cults	1690	272	40	29	488	721	21	17	1139	785	61	46	1627	120
	NC7-sun.wilds	2043	357	39	25	427	535	13	13	602	779	52	38	1029	356
	Total:	11108	3338	187	150	4096	2721	83	78	3839	5166	270	228	7935	1197
McCoy	NC7-medicinals	305	138	52	44	261	100	5	5	116	184	57	49	377	109
	Total:	305	138	52	44	261	100	5	5	116	184	57	49	377	109
Millard	NC7-corn.kin	34	6	7	7	11	0	0	0	0	6	7	7	11	2
	NC7-maize	18370	1755	345	243	4185	208	35	31	237	1826	380	274	4422	3131
	Total:	18404	1761	352	250	4196	208	35	31	237	1832	387	281	4433	3133
Reitsma	NC7-chicory	250	118	7	7	252	5	2	2	5	118	9	9	257	210
	NC7-cucumis.cucs	1351	1281	47	43	1703	1282	13	12	1646	1283	60	55	3349	317
	NC7-cucumis.melo	3106	574	42	39	919	237	17	16	252	738	59	55	1171	806
	NC7-cucumis.wilds	332	46	6	6	67	120	7	6	181	121	13	12	248	21
	NC7-cucurbita	986	825	48	46	1232	327	3	3	336	829	51	49	1568	151
	NC7-cucurbits.misc	2	1	2	2	2	0	0	0	0	1	2	2	2	1
	NC7-daucus	1087	345	20	20	431	59	3	3	60	375	23	23	491	590
	NC7-ocimum	96	85	23	21	204	0	0	0	0	85	23	21	204	17
	NC7-parsnips	71	1	1	1	1	0	0	0	0	1	1	1	1	15
	Total:	7281	3276	196	185	4811	2030	45	42	2480	3551	241	227	7291	2128
Widrlechner	NC7-mints	128	34	16	15	42	17	1	1	17	38	17	16	59	5
	NC7-ornamentals	1980	171	54	49	215	24	4	4	26	187	58	53	241	157
	Total:	2108	205	70	64	257	41	5	5	43	225	75	69	300	162
NCRPIS Total:		**47870**	**9731**	**746**	**553**	**14956**	**5582**	**169**	**146**	**7344**	**12289**	**915**	**699**	**22300**	**8655**

Year 2005 Table 3B

Internal NCRPIS Distributions

			NC7 Related (# Accs)							Seed Storage Maintenance	
CURATOR	GENUS CROP	# Accs	Backed Up	Germed	Obs	Regen	Path Test	Total	# Distinct Accs for NC7 Orders	# Accs Stored	# Accs Ct Rev
Brenner	NC7-amaranth	3330	68	567	71	100	0	806	711	86	85
	NC7-celosia	54	0	0	0	9	0	9	9	0	0
	NC7-echinochloa	302	27	223	48	23	0	321	268	0	11
	NC7-grasses	119	36	87	0	14	0	137	89	0	6
	NC7-legumes	227	0	0	0	1	0	1	1	0	20
	NC7-melilotus	952	17	27	0	25	0	69	52	28	19
	NC7-panicum	948	0	6	5	48	17	76	73	0	4
	NC7-perilla	22	0	0	1	0	0	1	1	0	0
	NC7-quinoa	234	0	0	1	7	0	8	8	0	0
	NC7-setaria	1006	0	255	1	0	88	344	338	0	2
	NC7-spinach	401	3	102	1	1	0	107	103	13	10
	NC7-umbels	1069	75	119	72	71	0	337	273	88	76
	Total:	8664	226	1386	200	299	105	2216	1926	215	233
Marek	NC7-asters	324	1	0	0	0	0	1	1	1	0
	NC7-brassica	2001	36	94	10	160	0	300	221	111	86
	NC7-crucifers	1197	55	64	0	310	0	429	350	72	48
	NC7-crucifers.pvp	1	0	0	0	0	0	0	0	0	0
	NC7-cuphea	650	3	5	5	34	0	47	41	4	49
	NC7-euphorbia	219	0	7	0	0	0	7	7	0	1
	NC7-flax	2818	0	0	2	12	0	14	14	0	13
	NC7-flax.wilds	165	6	9	3	72	0	90	87	15	2
	NC7-sun.cults	1690	24	17	0	53	36	130	120	56	112
	NC7-sun.wilds	24	0	0	0	0	0	0	0	0	0
	NC7-sun.wilds.ann	1340	31	23	69	48	2	173	150	34	98
	NC7-sun.wilds.per	679	87	90	2	104	0	283	206	129	51
	Total:	11108	243	309	91	793	38	1474	1197	422	460
McCoy	NC7-medicinals	305	54	65	35	57	43	227	109	79	5
	Total:	305	54	65	35	57	43	227	109	79	5
Millard	NC7-corn.kin	34	1	0	0	1	0	2	2	1	1
	NC7-maize	18370	76	1644	1480	345	359	3904	3131	530	2277
	Total:	18404	77	1644	1480	346	359	3906	3133	531	2278
Reitsma	NC7-chicory	250	0	0	210	0	0	210	210	0	1
	NC7-cucumis.cucs	1351	29	272	0	60	0	361	317	36	217
	NC7-cucumis.melo	3106	70	129	2	52	670	923	806	153	41
	NC7-cucumis.wilds	332	17	18	0	2	0	37	21	19	36
	NC7-cucurbita	986	18	128	0	39	0	185	152	29	42
	NC7-cucurbits.misc	2	0	0	0	0	0	0	0	0	0
	NC7-daucus	1087	52	529	19	71	0	671	590	119	217
	NC7-ocimum	96	0	4	0	14	0	18	17	0	71
	NC7-parsnips	71	15	15	3	0	0	33	15	15	3
	Total:	7281	201	1095	234	238	670	2438	2128	371	628
Widrlechner	NC7-mints	128	0	0	0	5	0	5	5	5	23
	NC7-ornamentals	1980	38	36	0	88	0	162	149	88	90
	Total:	2108	38	36	0	93	0	167	154	93	113
NCRPIS Total:		**47870**	**839**	**4535**	**2040**	**1826**	**1215**	**10428**	**8647**	**1711**	**3717**

Year 2005 Table 4 NCRPIS Accession (Acc) Observations (Obs) in GRIN, Images in GRIN

CURATOR	GENUS CROP	Number Accs	Number Acc Obs Trials	Number Acc Obs in Curator Notes	Number Obs in GRIN for Year	Number Acc Obs in GRIN for Year	Number Acc Obs in GRIN Last Year	Number Acc Obs in GRIN (all yrs)	Number Accs Imaged	Number Acc Images in GRIN for Year	Number Acc Images in GRIN (all years)
Brenner	NC7-amaranth	3330	71	124	1106	297	466	3310	111	34	251
	NC7-celosia	54	0	4	2	2	0	5	2	0	3
	NC7-echinochloa	302	48	51	263	52	10	291	44	0	0
	NC7-grasses	119	0	15	0	0	0	1	2	0	0
	NC7-legumes	227	0		0	0	0	84	1	0	0
	NC7-melilotus	952	0	5	2	2	0	912	4	0	0
	NC7-panicum	948	5	23	0	0	0	943	0	0	0
	NC7-perilla	22	1		0	0	0	0	0	0	1
	NC7-quinoa	234	1	1	10	10	0	229	0	0	1
	NC7-setaria	1006	1	1	0	0	0	993	1	0	0
	NC7-spinach	401	1	1	1	1	338	401	0	0	0
	NC7-umbels	1069	72	50	1	1	0	3	32	0	0
	Total:	8664	200	275	1385	365	814	7172	197	34	256
Marek	NC7-asters	324	0		0	0	0	4	1	0	0
	NC7-brassica	2001	10		497	117	82	1634	40	0	234
	NC7-crucifers	1197	0		137	33	85	582	101	1	223
	NC7-crucifers.pvp	1	0		5	1	0	1	0	0	0
	NC7-cuphea	650	5		0	0	0	326	1	0	0
	NC7-euphorbia	219	0		0	0	0	0	0	0	0
	NC7-flax	2818	2		12	12	3	2813	0	0	0
	NC7-flax.wilds	165	3		170	20	18	82	3	0	0
	NC7-sun.cults	1690	0		557	39	103	1616	0	1	1
	NC7-sun.wilds	24	0		0	0	0	15	0	0	0
	NC7-sun.wilds.ann	1340	69		348	44	101	1243	8	1	1
	NC7-sun.wilds.per	679	2		1318	99	15	464	25	0	0
	Total:	11108	91	0	3044	365	407	8780	179	3	459
McCoy	NC7-medicinals	305	35		6438	133	0	133	58	0	0
	Total:	305	35	0	6438	133	0	133	58	0	0
Millard	NC7-corn.kin	34	0		0	0	0	0	0	0	0
	NC7-maize	18370	1480		12187	2731	876	15393	3556	1444	3921
	Total:	18404	1480	0	12187	2731	876	15393	3556	1444	3921
Reitsma	NC7-chicory	250	210		571	250	0	250	0	0	0
	NC7-cucumis.cucs	1351	0	867	4373	1349	0	1349	52	0	0
	NC7-cucumis.melo	3106	2	850	8005	3094	0	3095	48	117	140
	NC7-cucumis.wilds	332	0	34	0	0	0	297	3	1	1
	NC7-cucurbita	986	0	442	2670	981	0	981	31	0	3
	NC7-cucurbits.misc	2	0		0	0	0	1	0	0	0
	NC7-daucus	1087	19	165	2707	1079	0	1079	53	0	0
	NC7-ocimum	96	0	18	291	96	0	96	6	0	0
	NC7-parsnips	71	3	3	0	0	0	0	0	0	0
	Total:	7281	234	2379	18617	6849	0	7148	193	118	144
Widrlechner	NC7-mints	128	0		0	0	0	0	5	0	0
	NC7-ornamentals	1980	8		56	48	0	79	57	48	48
	Total:	2108	8	0	56	48	0	79	62	48	48
NCRPIS Total:		**47870**	**2048**	**2654**	**41727**	**10491**	**2097**	**38705**	**4245**	**1647**	**4828**

CURATOR	GENUS CROP	TIME PERIOD	Number Orders	Number Recipients	Number Items Distributed	Number Accessions Distributed
Brenner	NC7-amaranth	01/01/2001 - 12/31/2001	47	36	908	596
		01/01/2002 - 12/31/2002	35	31	301	160
		01/01/2003 - 12/31/2003	43	42	921	516
		01/01/2004 - 12/31/2004	39	37	472	352
		01/01/2005 - 12/31/2005	59	53	540	229
	Total:		223	199	3142	1853
	NC7-celosia	01/01/2001 - 12/31/2001	2	2	12	11
		01/01/2002 - 12/31/2002	6	6	8	6
		01/01/2003 - 12/31/2003	3	3	17	14
		01/01/2004 - 12/31/2004	4	4	5	4
		01/01/2005 - 12/31/2005	5	5	7	6
	Total:		20	20	49	41
	NC7-echinochloa	01/01/2001 - 12/31/2001	4	4	35	32
		01/01/2002 - 12/31/2002	4	4	5	5
		01/01/2003 - 12/31/2003	5	5	26	26
		01/01/2004 - 12/31/2004	7	7	33	28
		01/01/2005 - 12/31/2005	5	5	59	57
	Total:		25	25	158	148
	NC7-grasses	01/01/2001 - 12/31/2001	3	3	7	6
		01/01/2002 - 12/31/2002	1	1	1	1
		01/01/2003 - 12/31/2003	1	1	1	1
		01/01/2004 - 12/31/2004	0	0	0	0
		01/01/2005 - 12/31/2005	1	1	3	3
	Total:		6	6	12	11
	NC7-legumes	01/01/2001 - 12/31/2001	3	3	6	6
		01/01/2002 - 12/31/2002	7	5	69	50
		01/01/2003 - 12/31/2003	5	5	31	26
		01/01/2004 - 12/31/2004	3	3	83	75
		01/01/2005 - 12/31/2005	7	6	28	24
	Total:		25	22	217	181
	NC7-melilotus	01/01/2001 - 12/31/2001	11	10	53	49
		01/01/2002 - 12/31/2002	5	5	43	42
		01/01/2003 - 12/31/2003	11	10	210	197
		01/01/2004 - 12/31/2004	9	7	68	58
		01/01/2005 - 12/31/2005	16	14	83	73
	Total:		52	46	457	419
	NC7-panicum	01/01/2001 - 12/31/2001	8	8	662	650
		01/01/2002 - 12/31/2002	2	2	9	9
		01/01/2003 - 12/31/2003	7	6	719	661
		01/01/2004 - 12/31/2004	9	8	920	877
		01/01/2005 - 12/31/2005	11	11	80	68
	Total:		37	35	2390	2265
	NC7-perilla	01/01/2001 - 12/31/2001	7	7	41	22
		01/01/2002 - 12/31/2002	4	4	26	22
		01/01/2003 - 12/31/2003	7	7	56	22
		01/01/2004 - 12/31/2004	3	3	21	14
		01/01/2005 - 12/31/2005	11	11	74	22
	Total:		32	32	218	102
	NC7-quinoa	01/01/2001 - 12/31/2001	18	15	239	173
		01/01/2002 - 12/31/2002	23	22	333	161
		01/01/2003 - 12/31/2003	22	20	275	195
		01/01/2004 - 12/31/2004	19	18	98	58
		01/01/2005 - 12/31/2005	32	30	302	138
	Total:		114	105	1247	725

	NC7-setaria	01/01/2001 - 12/31/2001	6	6	20	19
		01/01/2002 - 12/31/2002	9	8	48	43
		01/01/2003 - 12/31/2003	7	7	55	50
		01/01/2004 - 12/31/2004	12	12	132	117
		01/01/2005 - 12/31/2005	10	9	150	126
	Total:		44	42	405	355
	NC7-spinach	01/01/2001 - 12/31/2001	12	11	1736	354
		01/01/2002 - 12/31/2002	12	11	767	362
		01/01/2003 - 12/31/2003	14	12	321	260
		01/01/2004 - 12/31/2004	15	14	80	71
		01/01/2005 - 12/31/2005	12	12	399	370
	Total:		65	60	3303	1417
	NC7-umbels	01/01/2001 - 12/31/2001	13	12	98	93
		01/01/2002 - 12/31/2002	21	17	294	208
		01/01/2003 - 12/31/2003	17	15	248	150
		01/01/2004 - 12/31/2004	33	32	353	193
		01/01/2005 - 12/31/2005	27	25	239	215
	Total:		111	101	1232	859
	Brenner Total:		754	693	12830	8376
Marek	NC7-asters	01/01/2001 - 12/31/2001	4	4	8	7
		01/01/2002 - 12/31/2002	7	5	14	11
		01/01/2003 - 12/31/2003	7	7	25	21
		01/01/2004 - 12/31/2004	4	4	7	6
		01/01/2005 - 12/31/2005	9	9	73	70
	Total:		31	29	127	115
	NC7-brassica	01/01/2001 - 12/31/2001	34	31	459	408
		01/01/2002 - 12/31/2002	51	47	527	361
		01/01/2003 - 12/31/2003	57	49	1562	795
		01/01/2004 - 12/31/2004	57	50	4347	1735
		01/01/2005 - 12/31/2005	56	54	1688	1286
	Total:		255	231	8583	4585
	NC7-crucifers	01/01/2001 - 12/31/2001	20	17	634	265
		01/01/2002 - 12/31/2002	24	23	241	212
		01/01/2003 - 12/31/2003	15	15	89	79
		01/01/2004 - 12/31/2004	31	27	1403	793
		01/01/2005 - 12/31/2005	52	48	1486	497
	Total:		142	130	3853	1846
	NC7-crucifers.pvp	01/01/2001 - 12/31/2001	0	0	0	0
		01/01/2002 - 12/31/2002	0	0	0	0
		01/01/2003 - 12/31/2003	0	0	0	0
		01/01/2004 - 12/31/2004	0	0	0	0
		01/01/2005 - 12/31/2005	0	0	0	0
	Total:		0	0	0	0
	NC7-cuphea	01/01/2001 - 12/31/2001	16	12	712	481
		01/01/2002 - 12/31/2002	10	8	247	216
		01/01/2003 - 12/31/2003	19	12	389	244
		01/01/2004 - 12/31/2004	14	10	229	180
		01/01/2005 - 12/31/2005	20	13	451	277
	Total:		79	55	2028	1398
	NC7-euphorbia	01/01/2001 - 12/31/2001	1	1	1	1
		01/01/2002 - 12/31/2002	4	4	8	6
		01/01/2003 - 12/31/2003	2	2	9	7
		01/01/2004 - 12/31/2004	1	1	3	2
		01/01/2005 - 12/31/2005	1	1	1	1
	Total:		9	9	22	17
	NC7-flax	01/01/2001 - 12/31/2001	13	13	267	223
		01/01/2002 - 12/31/2002	8	8	73	63
		01/01/2003 - 12/31/2003	6	6	96	95
		01/01/2004 - 12/31/2004	16	15	211	201
		01/01/2005 - 12/31/2005	13	13	1546	1441
	Total:		56	55	2193	2023

	NC7-flax.wilds	01/01/2001 - 12/31/2001	2	2	22	19
		01/01/2002 - 12/31/2002	2	2	18	12
		01/01/2003 - 12/31/2003	3	3	20	19
		01/01/2004 - 12/31/2004	6	6	69	32
		01/01/2005 - 12/31/2005	6	6	34	30
	Total:		19	19	163	112
	NC7-sun.cults	01/01/2001 - 12/31/2001	41	30	1434	759
		01/01/2002 - 12/31/2002	44	36	562	422
		01/01/2003 - 12/31/2003	43	34	543	405
		01/01/2004 - 12/31/2004	38	31	310	210
		01/01/2005 - 12/31/2005	61	46	1627	785
	Total:		227	177	4476	2581
	NC7-sun.wilds	01/01/2001 - 12/31/2001	32	27	1259	858
		01/01/2002 - 12/31/2002	32	20	890	652
		01/01/2003 - 12/31/2003	34	22	473	355
		01/01/2004 - 12/31/2004	44	35	549	386
		01/01/2005 - 12/31/2005	52	38	1029	779
	Total:		194	142	4200	3030
	Marek Total:		1012	847	25645	15707
McCoy	NC7-medicinals	01/01/2001 - 12/31/2001	18	18	356	88
		01/01/2002 - 12/31/2002	22	17	207	96
		01/01/2003 - 12/31/2003	35	27	387	122
		01/01/2004 - 12/31/2004	31	29	221	112
		01/01/2005 - 12/31/2005	57	49	377	184
	McCoy Total:		163	140	1548	602
Millard	NC7-corn.kin	01/01/2001 - 12/31/2001	7	7	13	7
		01/01/2002 - 12/31/2002	5	5	16	7
		01/01/2003 - 12/31/2003	7	7	22	8
		01/01/2004 - 12/31/2004	8	8	11	6
		01/01/2005 - 12/31/2005	7	7	11	6
	Total:		34	34	73	34
	NC7-maize	01/01/2001 - 12/31/2001	270	198	2637	1606
		01/01/2002 - 12/31/2002	399	279	4714	2511
		01/01/2003 - 12/31/2003	226	178	2298	1475
		01/01/2004 - 12/31/2004	334	241	4473	2207
		01/01/2005 - 12/31/2005	380	274	4422	1826
	Total:		1609	1170	18544	9625
	Millard Total:		1643	1204	18617	9659
Reitsma	NC7-chicory	01/01/2001 - 12/31/2001	6	6	288	175
		01/01/2002 - 12/31/2002	8	8	261	134
		01/01/2003 - 12/31/2003	8	7	192	144
		01/01/2004 - 12/31/2004	5	4	45	43
		01/01/2005 - 12/31/2005	9	9	257	118
	Total:		36	34	1043	614
	NC7-cucumis	01/01/2001 - 12/31/2001	59	49	1229	933
		01/01/2002 - 12/31/2002	58	46	2658	1773
		01/01/2003 - 12/31/2003	46	36	1901	1391
		01/01/2004 - 12/31/2004	73	64	1393	1105
		01/01/2005 - 12/31/2005	101	89	4768	2142
	Total:		337	284	11949	7344
	NC7-cucurbita	01/01/2001 - 12/31/2001	22	20	288	156
		01/01/2002 - 12/31/2002	20	17	165	132
		01/01/2003 - 12/31/2003	11	11	170	150
		01/01/2004 - 12/31/2004	38	35	702	490
		01/01/2005 - 12/31/2005	51	49	1568	829
	Total:		142	132	2893	1757

	NC7-cucurbits.misc	01/01/2001 - 12/31/2001	2	2	2	1
		01/01/2002 - 12/31/2002	0	0	0	0
		01/01/2003 - 12/31/2003	1	1	1	1
		01/01/2004 - 12/31/2004	1	1	2	2
		01/01/2005 - 12/31/2005	2	2	2	1
	Total:		6	6	7	5
	NC7-daucus	01/01/2001 - 12/31/2001	13	12	235	211
		01/01/2002 - 12/31/2002	11	11	75	67
		01/01/2003 - 12/31/2003	13	12	426	294
		01/01/2004 - 12/31/2004	21	21	596	378
		01/01/2005 - 12/31/2005	23	23	491	375
	Total:		81	79	1823	1325
	NC7-ocimum	01/01/2001 - 12/31/2001	4	4	96	79
		01/01/2002 - 12/31/2002	7	7	18	16
		01/01/2003 - 12/31/2003	8	8	52	42
		01/01/2004 - 12/31/2004	22	22	235	84
		01/01/2005 - 12/31/2005	23	21	204	85
	Total:		64	62	605	306
	NC7-parsnips	01/01/2001 - 12/31/2001	0	0	0	0
		01/01/2002 - 12/31/2002	2	2	9	8
		01/01/2003 - 12/31/2003	1	1	1	1
		01/01/2004 - 12/31/2004	3	3	5	4
		01/01/2005 - 12/31/2005	1	1	1	1
	Total:		7	7	16	14
	Reitsma Total:		673	604	18336	11365
Widrlechner	NC7-mints	01/01/2001 - 12/31/2001	5	5	75	42
		01/01/2002 - 12/31/2002	4	4	22	19
		01/01/2003 - 12/31/2003	8	8	44	38
		01/01/2004 - 12/31/2004	17	16	45	28
		01/01/2005 - 12/31/2005	17	16	59	38
	Total:		51	49	245	165
	NC7-ornamentals	01/01/2001 - 12/31/2001	46	42	205	132
		01/01/2002 - 12/31/2002	56	50	350	259
		01/01/2003 - 12/31/2003	81	72	496	198
		01/01/2004 - 12/31/2004	87	81	361	220
		01/01/2005 - 12/31/2005	58	53	241	187
	Total:		328	298	1653	996
	Widrlechner Total:		379	347	1898	1161
NCRPIS Total:			**4624**	**3835**	**78874**	**46870**

Year 2005 Table 6 **NC7 CSREES Region Order History**

TIME PERIOD	Total Number of Orders	Number of Orders (DI)	Foreign Orders (DI)	Domestic Orders (DI)	Domestic Orders (DI) CSREES Regions				
					NC7	NE9	S9	W6	APO
01/01/2005 to 12/31/2005	1220	915	169	746	341	76	197	132	
01/01/2004 to 12/31/2004	1044	807	164	643	303	74	159	107	
01/01/2003 to 12/31/2003	871	637	157	480	221	53	100	106	
01/01/2002 to 12/31/2002	1034	782	172	610	305	71	133	101	
01/01/2001 to 12/31/2001	862	651	166	485	241	51	91	102	
01/01/2000 to 12/31/2000	781	549	149	400	205	46	74	75	
01/01/1999 to 12/31/1999	817	576	174	402	188	54	78	81	1

North Central Regional Plant Introduction Station Staff