

NCRPIS ANNUAL REPORT - 2009

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**NORTH CENTRAL REGIONAL PLANT INTRODUCTION STATION
NC-7 ANNUAL REPORT, JANUARY 1 - DECEMBER 31, 2009**

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

Voting members

1. Illinois	G. Kling	7. Missouri	J. Shannon
2. Indiana	J. Janick	8. Nebraska	D. Santra
3. Iowa	R. Hall	9. N. Dakota	B. Johnson
4. Kansas	M. Stamm	10. Ohio	D. Francis
5. Michigan	A. Iezzoni	11. S. Dakota	K. Glover
6. Minnesota	J. Orf	12. Wisconsin	W. Tracy

Non-voting members

13. California-Davis	R. Karban	18. Michigan	J. Hancock
14. Connecticut	M. Brand	19. Missouri	P. Bueselinck
15. Delaware	J. Hawk	20. New Jersey	T. Molnar
16. Illinois		21. Texas	D. Baltensperger
17. Iowa	K. Lamkey	22. Wisconsin	S. Kaeppler
18. Kansas	A. Fritz	23. Wisconsin	N. de Leon

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	L. Chandler
4. Cooperative State Research, Education and Extension Service	A. Thro
5. National Center for Agric. Util. Research	*T. Isbell
6. National Center for Genetic Resources Preservation	*D. Dierig

*Voting members

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

See organizational chart, Figure 1 in the Appendix.

III. PROGRESS OF WORK AND PRINCIPAL ACCOMPLISHMENTS:

Personnel changes - June, 2009– May, 2010:

Departures:

Barbara Bingaman, ISU Agricultural Specialist, July, 2009

New Hires:

Bruce Hall, Agricultural Research Science Technician, Maize, May, 2010

Promotions:

Pete Cyr, Applications Software Dvlp. IT Specialist, to GS-13, May, 2010

Management of Federal STEP (Student Temporary Employees):

USDA-ARS resources provided for 17 student labor positions in FY09, a decrease from FY09. We limited equipment purchases, postponed recovering a greenhouse for a second year and focused on increasing the availability of the collections and quality of associated information. The STEP positions support curatorial activities including regeneration, seed processing, viability testing, farm and facilities operations, and IT support. Students were interviewed and selected by ISU Program Manager Larry Lockhart or ARS maize technicians Matt Lively and Irvin Larsen. Marci Bushman and Rachael Beyer managed the administrative aspects of all STEP hires, with support and guidance by Ames ARS HR Specialist Kim Grandon and Admin. Officer Carol Moran. The NIH grant provided for 0.5 STEP, and a USDA/DOE Biomass grant provided for employment of one ISU student employee.

Budget:

Declining purchasing power of budgets and rising salary, energy and operational expenses continue to erode our ability to support core functions. In FY09-11, Hatch funds used for ISU staff salaries and some expenses are being supplemented by a component of a USDA/DOE grant evaluating *Helianthus argophyllus* for biomass potential on which Oilseeds Curator Dr. Laura Marek serves as a co-PI with PI Steve Knapp, Univ. of Georgia. The ARS PIRU Unit's medicinal curator position and a student position are supported 50% by ARS Horticulturist Dr. Mark Widrlechner's NIH research grant sub-contract on Botanical and Dietary Supplements through March of 2011, for which he serves as co-PI with Dr. Diane Birt of Iowa State University. Dr. Charles Block's Sclerotinia Initiative funding supports student labor and supplies used in that research. GRIN-Global grant funds support the salaries of contract development personnel and project connected travel of ARS personnel.

Construction and Facilities:

Heaters were replaced in Greenhouses #1 and #3. Old ballasts were replaced with electronic ballasts to improve energy efficiency. Energy efficient Point of Service water heaters were installed in the shop and headhouse. A kitchen was developed in the HQ building after dividing an existing room in half. A survey was completed for lease of the bee overwintering structure and greenhouse #3 by ARS, which will enable either NC7 or ARS federal resources to be used for maintenance in the future. In FY11, we plan to recover Greenhouse #3.

Equipment:

Major equipment purchases were facilitated by the ARRA, which provided funds to update six vehicles in the fleet, including a flatbed truck with lift to replace a truck assigned to the pollinator project. Other purchases included a 25 Hp tractor to replace an aging Kubota, a new band saw for the machine shop, a Uni-Hydro Iron Worker for the shop, and 140 sunflower cage frames to support regenerations (this item funded by the USDA-ARS Office of National Programs).

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

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

Acquisition and Documentation Highlights:

In 2009, (Appendix Table 1) 678 accessions were acquired, equal to 1.3% of the previous collection holdings. Of these, 521 were received from within the NPGS through exploration and transfer. This compared with 564 new accessions in 2008, 450 in 2007, and 650 in 2006. Newly acquired germplasm included the first accessions brought into the NCRPIS under the Standard Material Transfer Agreement (SMTA), 29 teosinte collections from CIMMYT in Mexico. In addition, there were 39 expired PVP lines, 24 GEM line releases, and six TZ inbreds that passed through quarantine. *Helianthus* acquisitions included 18 cultivated *H. annuus* with expired CSR property rights, via the NCGRP; 18 *H. argophyllus* from Australia; 51 wild populations from a National Plant Germplasm System (NPGS) Plant Exploration Office (PEO) funded exploration of Kansas, Oklahoma, Arkansas, and Missouri; five wild populations from a collaborator in Kansas; and additional accessions. New vegetable accessions include 91 *Daucus* collected in Tunisia by PEO- funded exploration; 10 *Cucumis sativus* cultivars used as disease differentials from the NCGRP, and heirloom cultivars. The horticulture project received 245 new accessions of ornamentals and mints, many resulting from transfer of collections made by the Department of Interior's Seeds of Success (SOS) program in the Western U.S.; *Fraxinus* collections made in Illinois, Wisconsin, and China, and funded by the PEO; and wild *Aronia* populations collected and donated by NC7 participant Mark Brand, University of Connecticut. In addition, 37 Umbelliferae were acquired, 29 from the exploration in Tunisia; one accession of federally threatened and endangered *Amaranthus pumilis* from Delaware; miscellaneous grasses acquired via the SOS program; *Chenopodium* and *Marina parryi* via Bureau of Land Management collections; Russian and Tunisian *Melilotus*; and a wild spinach relative, *Monolepis nuttalliana* from Iowa. Forty new accessions were acquired of medicinal species, bringing the collection total to 489 accessions. These included *Hypericum* and *Prunella* native populations from southern Wisconsin, northern Illinois and Ohio, and five *Prunella* accessions from Asia.

Of ongoing concern is the successful entry of germplasm collected from international explorations into the U.S. It is critical that clean, pest- and pathogen-free seed be shipped or carried in by collectors; sufficient time needs to be devoted to collection

sample preparation and sufficient care post-collection. Excellent quantities of seed provided by collectors of many new accessions have made a significant proportion available and distributable immediately.

Ninety five accessions were assigned permanent PI numbers in 2009. Taxonomic re-identification was completed for 75 accessions; 215 accessions were nominated for inactivation, 10 of these due to duplication. R. Stebbins continues to enter old passport information from logbooks for early Ames-numbered accessions.

Original seed samples continue to be scanned by L. Pfiffner and L. Burke, in order to provide useful visual references for comparison of regeneration lots with original samples.

Regeneration and Maintenance Highlights:

In 2009, 1,096 accessions were grown for regeneration and 1,017 were harvested, as compared to 1,443 accessions grown for regeneration and 1,113 harvested in 2008. An additional 123 perennials are growing in permanent plantings. Over 1,040 accessions were made available to the public. Accessions backed up at the NCGRP in Ft. Collins in 2009 numbered 1,848; this compares with 499 lots in 2008, 2970 lots in 2007, and 880 in 2006. Seventy nine percent of the NCRPIS collections are backed up (Appendix Table 2). Overall collection available is 74%, the same as in 2009, despite 3% growth in collection size since 2006. An additional 1,545 accessions were sent to Ft. Collins for assembly with accessions from other NPGS sites and deposit to the Svalbard Global Germplasm Vault.

Using an application designed in-house, Pocket Pollinator, information on pollinator insect actions connected with regeneration efforts is collected by our entomology staff and entered into the GRIN database. Over time, this will provide a rich database that can be explored to better understand effective plant-pollinator interactions with respect to seed production.

Assistance in regeneration was provided by USDA-ARS staff of Parlier, CA for increase of wild *Helianthus* taxa. *Daucus* regeneration efforts were supported by seed increases from Seminis Vegetable Seeds (R. Maxwell) and Nunhems (R. Freeman). USDA-ARS staff of Mayaguez, PR and the St. Croix quarantine nursery staff supported regeneration of 116 maize accessions. GEM Project Coordinator Michael Blanco provided resources in Puerto Rico to increase 10 tropical inbreds, as the maize curation project did not have the resources for a winter nursery in 2009, and University of Wisconsin graduate student Tony Studer provided pollination assistance. Syngenta provided resources for increase of 14 tropical accessions on Kauai.

In addition to the *Daucus* regenerations grown in Ames, seed increases were received from R. Maxwell, Seminis Vegetable Seeds, ID, and R. Freeman, Nunhems, OR. Dr. Freeman and Dr. Maxwell were sent additional PI-numbered accessions, for regeneration in 2009. Spinach regenerations were supported by cooperative efforts between the USDA-ARS and Sakata Seed America, Inc. in Salinas, CA.

Distribution:

2009 external distributions included 26,904 items of 13,515 unique accessions to fulfill 1,487 orders from 1081 requestors. This compares with 2009 distributions of 24,726 items of 12,709 unique accessions to fulfill 1,243 orders from 908 individuals, exceeding the previous record high (2006) of 26,100 items of 13,789 accessions. Approximately 20% were distributed internationally, and 80% to domestic researchers (Appendix Table 3A). A detailed listing of distributions by site crop can be found in Appendix Table 5. The relative numbers of distributions generally correlate well with the proportional makeup of the collections, although demand for maize demand is always larger than for other crops. An additional 8,971 items were distributed within the NCRPIS for all internal genebank activities (Appendix Table 3B).

Curator	Collection Size – 2009	% of Total Collections	% of 2009 Ext. Distributions
Brenner	8932	18	12
Marek	11262	22	16
Qu	489	<1	<1
Millard	20200	40	43
Reitsma	7545	15	25
Widrlechner	2490	5	3
Totals	50313	100	100

Research demand for our plant genetic resources collections continues to be very high; requests for biofuel applications and health and nutrition contribute increasingly to these increases, as well as for basic research applications, disease and insect resistance, photoperiod response, and an array of performance traits. *Echinacea* and *Prunella* plant materials were provided for NIH-funded research on botanical dietary supplements at ISU.

NPGS curators at all sites continue to receive many requests from individuals not affiliated with research institutions, generally for home gardening. Home gardeners are redirected to other sources of commercially available materials. Although our resources cannot support maintaining and distributing the collections to home gardeners, we inform these requestors about plant genetic resource conservation, and encourage interested individuals to save seeds, conserve them, and share germplasm and associated information. The proliferation of websites instructing non-research requestors how to deceive curators at various germplasm sites in order to get free germplasm is problematic. The careful efforts that go into each and every increase, characterization, imaging, processing, storage, viability testing, and distribution surely make these seeds among the most expensive to provide in the world.

Evaluation and Characterization:

In 2009, the NCRPIS utilized 7,874 accessions for observation, evaluation and characterization for a wide array of descriptor information. About 15,700 observations were entered in the GRIN database (<http://www.ars.grin.gov/npgs/>), less than half the amount of 2008. Images added to GRIN for the year number 589 (Appendix Table 4).

The amaranth project (Brenner & Flomo) loaded over 1,450 *Melilotus* observation from previous regenerations dating back to 1990. Two maize disease screening nurseries were evaluated by Pioneer-Hi-Bred/DuPont for northern leaf blight and Diplodia ear rot. NCRPIS Pathologist Block screened 298 accessions for Stewart's wilt resistance. The maize project loaded over 10,200 observations. The vegetable project's *Daucus* and *Cichorium* evaluation projects have generated large descriptor datasets. Our oilseeds curation and pathology teams and USDA collaborators at Fargo screened *Helianthus* accessions for Sclerotinia resistance at multiple locations in 2009, including the best 20 accessions from 2008 testing. Field testing results correlated well with greenhouse screening results.

Information technology and telecommunications:

The NCRPIS is providing expertise and leadership for the development of GRIN-Global (the successor to the GRIN system); this has become the primary focus of two NCRPIS staff members. This project is undertaken as a partnership between USDA-ARS, Bioversity International and the Global Crop Diversity Trust (the Trust) to develop a genebank information management system which can be deployed to any genebank in the world. National Program 301 Leader, Peter Bretting is the PI for this agreement.

The basic GRIN database schema is recognized as essential to preserve; improved interfaces and views are required to support the needs of genebank curatorial personnel and external, public users of information associated with the collections. The source code for the products of development will be freely available. The system will be able to function on either networked systems or stand-alone PCs, and will be free of recurring licensing costs.

Ames-based development team members include Pete Cyr, NCRPIS Applications Software Development IT Specialist, Project Manager; Mark Millard, NCRPIS Maize Curator, Analyst; Lisa Burke, NCRPIS Seed Storage Manager, beta tester. Other Ames-based personnel include developer Brock Weaver and web developer Rachelle Little, Bioversity consultants. In 2009, Joseph Postman, Pathologist at the National Clonal Germplasm Repository in Corvallis, OR, and Tomás Ayala-Silva of the Miami Sub-Tropical Research Station began providing valuable input as testers representing the clonal germplasm community. An EMBRAPA scientist from Brazil, Patrícia Goulart Bustamante, spent six weeks in Ames evaluating GRIN-Global for potential use by EMBRAPA, and contributing to system development. Together with the extensive work of the staff of the USDA-ARS Database Management Unit (DBMU) in Beltsville, MD, responsible for the current GRIN system, representatives of seed- and clonally-propagated NPGS sites, and our international partners, they are working to accomplish ambitious project objectives within tight timelines. U.S. curators and Bioversity International personnel tested a GRIN-Global System release candidate (RC1) in December 2009. Work on the new public GRIN-Global interface progressed in 2009; it will be deployed in late 2010.

Please see IT section for technically detailed reports on support activities.

Germplasm's Viability and Health:

Over 2,530, or 5% of the NCRPIS collections, were tested for viability in 2009, fewer than in the past due to labor constraints (Appendix Table 2).

Experimental viability testing methods to release seed dormancy in *Echinochloa* and *Actaea* were explored. The progress of after-ripening in *Calendula* was studied with a goal of understanding loss of seed dormancy in cold storage. Alternative treatments to six week cold-stratification treatments were explored for breaking seed dormancy of wild *Helianthus*. Use of a thermal gradient table has enabled D. Kovach to conduct a series of experiments comparing controlled temperature, light and humidity conditions to establish appropriate seed germination testing protocols for various crops. The table also is used by graduate student Ivan Ayala Diaz in his studies of *Thlaspi* and *Camelina*.

Pathology team research (C. Block), focused on combining greenhouse and field resistance screening methods for Sclerotinia stalk rot in wild sunflowers; long-term survival and seed transmission of bacterial fruit blotch (*Acidovorax avenae* subsp. *citrulli*), or BFB, in Cucurbits with an emphasis on identifying infected, older *Cucumis melo* seedlots; and regular disease monitoring of cucurbit plantings from transplant to harvest. Screening of the entire 2300 accessions of the melon collection for BFB is now completed. Field observations were made in the increase plots, and accessions were monitored in particular for diseases for which seedborne transmission is of concern.

Insect management:

Entomologists S. Hanlin and S. McClurg supplied 640 accessions in 696 cages with five types of pollinator insects in 2009. Detailed, interesting observations and interpretative information regarding their field pollinator research activities can be found in their extensive section of the annual report for information on their continuing efforts to enhance the pollination program's effectiveness and efficiency, and many interesting observations. Substantial report space is devoted to this team's activities because of the uniqueness of this project, and relevance to the broader germplasm conservation world. Feedback and suggestions on experimental approaches are welcomed.

The staff continues to compare use of solid 'fondant' sugar with corn syrup for feeding the honey bees. While fondant sugar use decreases labor needs, honeybees cannot store this sugar as a resource for winter. During the summer months, food source did not impact nucleus hive strength. It is necessary to use corn syrup feed in the early spring to dispense medication and in the late summer to enable the bees to build honey stores for overwinter survival.

We continue to consider the impact of the effectiveness of insect pollinators on cross-fertilization of caged plantings, and whether the genetic profile of the accession is maintained during regeneration. This will be a focus of future studies, and will require careful selection of parent lots and assay of their progeny using molecular markers or known discernible traits.

Enhancement:

The Germplasm Enhancement of Maize Project, or GEM, continues to work with public and private collaborators to adapt exotic maize germplasm to broaden the genetic diversity of temperate U.S. maize production and provide unique, key priority traits. Research and breeding is designed to identify traits and genes to

support improvement of agronomic productivity, disease resistance, insect resistance, and value-added grain characteristics, including total extractable starch to support ethanol production, and resistant starch – of importance to human health and nutrition.

The Project has released 212 lines from 2001-2009, representing over 40 maize races. An important goal is development of a set of inbred lines representative of the diversity inherent to all of the races of maize. In addition to traditional introgression methods, the project is using a haploid inducer line and generating double haploid maize lines. With the collaboration of AgReliant and Monsanto, our 400 initial double haploid seeds were increased in Hawaii and Chile in winter 2009. The ISU Dihaploid Facility is an important partner in this effort.

Photoperiod sensitive tropical maize often does not flower until September in Ames. GEM and maize curation teams have continued to collaboratively develop an effective method for photoperiod control in the field. While successful, it is difficult to achieve the field scale needed to support the number of accessions that require photoperiod control treatment. This effort has been leveraged by the sunflower project, which has used it very effectively to induce flowering in certain wild sunflower accessions. Photoperiod-control environment capacity on the order of one to three acres would be useful.

A collaborative project to evaluate outcrossing of grain amaranth cultivars with weedy amaranth species is in progress.

Outreach and Scholarship:

Approximately 305 visitors toured the NCRPIS during 2009, including the extended stay of a senior EMBRAPA researcher, and the GRIN-Global Technical Steering Group. 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:

Processes involved in regeneration, characterization, and making viable germplasm available are labor intensive. Currently, resources do not allow maintenance and regeneration efforts, including viability testing, to keep pace with demand. We will continue to try to improve conservation methods to better use the resources available to us, and to develop labor and resource saving technologies. We continue to evaluate activities that can be reasonably reduced without sacrificing collection health and quality, and to improve efficiency.

Continued emphasis will be placed on communicating with research stakeholders to identify and address collection development needs. Crop collections for biofuels and medicinal/nutriceutical applications need to be enhanced; wise selection of targets for these efforts requires use of complex and varied sources of information. In 2010, collection expeditions are planned to acquire *Helianthus* from the southern and south central U.S., *Fraxinus* from the central U.S., wild spinach relatives from the

High Plains; and *Chenopodium* and *Amaranthus*. *Thlaspi* acquisition is anticipated via collaborators.

Better characterization information is essential to enable well-target use of the collections, especially given the increasing constraints of limited research and conservation resources. Collaboration between vegetable curation staff at the NCRPIS and ARS researchers in Wisconsin will focus on *Daucus* characterization and taxonomy in 2010. A major effort to phenotypically and genomically characterize the entire maize inbred collection will be conducted in 2010. Oilseeds curation staff will increase the *Thlaspi* and *Camelina* collections in order to better support biofuel researchers.

Horticulturist 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. The project is complete, save for public distribution. In addition, he serves as an investigator on an NIH grant to develop collections of medicinal plants and elucidate the basis of the phytopharmaceutical activity, and is working to develop inter-agency coordination of *Fraxinus* collection in the face of the Emerald Ash Borer threat. Curator L. Marek serves as a co-PI on a USDA/DOE grant evaluating woody biomass traits in sunflower. In addition to extensive phenotypic data capture, and providing germplasm seeds and tissues in support of the genetics and genomics project aspects, extensive images were captured in 2009; extensive resources will be committed in 2010 as well.

Software development efforts for the next two years will center on the development and deployment of the successor to the GRIN system, GRIN-Global - its schema, internal and public interfaces. These efforts are facilitated by contributions from germplasm stakeholders in the U.S. and abroad, as we seek examples of use cases and desired features and functionalities of the new system.

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 Appendix Table 6 for a summary of the various CSREES (now NIFA) regions' order history, illustrating the demand for plant genetic resources to support research and educational activities. NC7 Region researchers typically account for nearly half of domestic plant germplasm distributions. 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.

The linkage of the GEM Project, the maize curation project, and public and private collaborators throughout the U.S. has resulted in synergy which facilitates 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, whom 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 of 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 2009 Annual Meeting include the following:

- Guidance was provided for development of the NC-7 mid-cycle review.
- The 2009 TAC meeting hosted by the NCRPIS in Ames, IA highlighted the extensive investigations of NC-7 participants using plant genetic resources to explore new agricultural products and new cropping systems. The

opportunities afforded by the meeting and field tours are key to establishing the types of collaborative relationships that lead to long-term partnerships for major research and development efforts.

- 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.
- Clarity on correct and consistent access to germplasm resources is needed and appreciated.
- The NC-7 RTAC supports the efforts of the ASTA and other stakeholders to actively pursue doubling of federal funding available to support this project and maintain its operations at the current level.
- 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 (repeat from 2007).
- The public GRIN interface should be significantly improved for ease of use and information delivery (repeat from 2007).
- Insufficient numbers of individuals are being trained in plant breeding and genetics, plant pathology, entomology, and related disciplines to meet current and future US needs; this is of serious concern (repeat from 2007).

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 19.0 FTE's. We coordinated and completed facility construction and upgrades.

Labor:

During 2009, 100 applications for hourly employment were received and reviewed. There were 47 interviews, resulting in 42 new or returning hourly employees hired. Currently there are 32 Biological Science Aides (13.3 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. He is now working half-time for the farm support group and half-time for the oilseeds project. The change has been very positive for both groups.

Brian Buzzell (Farm Mechanic) joined the staff in May 2002.

Scott McCubbin (STEP) was shared with the pollination project.

Maintenance projects:

During the past year the farm staff initiated and completed several projects which enhanced the efficiency and safety of the station operations. Budget limitations this past year have impacted the number and scope of projects completed by the farm staff.

1. Replaced one greenhouse heater in GH1 and two in GH3
2. Replaced several old ballasts with electronic ballasts to improve energy efficiency
3. Installed energy efficient Point of Service water heaters in shop and head house
4. Completed kitchen in headquarters building
5. Completed survey for lease of bee overwintering and greenhouse #3 buildings

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. Flatbed truck with lift to replace truck assigned to Pollinator Project
2. Twenty-five Hp tractor to replace aging Kubota
3. 140 Sunflower Cage Frames
4. Band saw for machine shop
5. Gator Utility Vehicle
6. Uni-Hydro Iron Worker

Tours:

This past year, we organized and conducted 17 tours. There were approximately 305 visitors to the NCRPIS during 2009.

Staff Training:

We conducted Tractor and Utility Vehicle Safety, Worker Right-to-Know and Worker Protection Standard training sessions for the new staff and student employees as well as updates for existing staff.

B. Information Technology and Telecommunications (P. Cyr and J. Perrett)

Jesse Perrett has been acting as the first-line of support for NCRPIS during 2009. Jesse was supervised by Pete Cyr who has been temporarily re-assigned to manage a 3-year project to rewrite the GRIN Germplasm Management System. The following list outlines the progress made by the IT team during 2009 at NCRPIS.

Equipment:

As of December 2009 NCRPIS has 64 workstations installed for use by permanent staff members and part-time temporary student help. In 2009, 18 workstations were deployed to replace aging equipment on curatorial staff desktops. Ten of the workstations were new Dell Optiplex 745 and 755's. Eight older machines were replaced with Optiplex 280's. 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. Thirty-eight end of life computers were surplus.

NCRPIS upgraded to a VLAN set up between all campus computers and the isolated network on the farm. This isolated network tunnel alleviates multiple synchronization problems and dual VPN concerns as well as increasing productivity for campus personnel. A new custom-designed server backup system was installed to replace the old system. The new system still uses Retrospect Multi Server, but is capable of performing backups faster and more reliably with less power use and the ability to shut down the drives when not in use. The system now uses SATA drives versus old IDE in order to lower cost and increase performance and usability. Forty desktop and laptop computers received 4GB memory upgrades to improve speed and reliability. The door security system's wireless access point to one of the greenhouses was replaced, and the entire system was upgraded to provide enhanced security and reliability. A new Epson Expression 10000XL scanner was installed and configured for life-cycle replacement. Six aging point-and-shoot digital cameras were replaced with Canon Digital Rebel XSi digital SLRs in order to improve archival image quality. One Dell XT2 tablet and one Sony Vaio P were implemented in order to improve field data collection. Four users' computers were upgraded to new Laptops including replacing the setup for one user who had two separate desktop machines with one laptop and two docking stations.

Software:

All of the workstations at NCRPIS are standardized on Windows XP with Service Pack 3 installed for increased security and reliability. Frequent updates to anti-

virus definitions and anti spy-ware definitions help ensure that these workstations remain vulnerability free.

During 2009, all workstations and servers at NCRPIS received security updates from Microsoft every month (on the second Tuesday of the month). PatchLink software was used to manage, track, and apply non-Microsoft software vulnerability patches for all non-Microsoft software in use on the workstations and servers at NCRPIS.

All computer systems on campus and at the farm (servers and workstations) use Symantec Endpoint Protection for enhanced security against virus and spyware threats.

A new VPN system for ARSNet connectivity was implemented in order to mitigate security concerns. The desktop per user VPN was required for about 5 months in order to check email, and is now used to load images to the GRIN database. All compatible laptop systems were encrypted using McAfee Endpoint Encryption whole hard drive encryption software. The TrackIt help desk software and Retrospect Multi Server Backup software were upgraded to new versions. Windows 7 and Server 2008 testing were performed in order to be ready for future upgrades.

Documentation:

IT support videos and training documents, and information about farm operation, safety, and health were posted to the NCRPIS intranet website. Regular input was provided to the area IT office regarding system and component information for data calls including information about: laptops, enterprise architecture, wireless networking equipment, privileged accounts, and video conferencing equipment.

Plans for 2010:

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

Upgrade intranet site to Microsoft Office SharePoint Server 2007 using SQL Server 2008 as the database.

Implement ThumbsPlus system for image archival and indexing on DVDs.

Replace Firebox with Cisco Adaptive Security Appliance.

Begin upgrading Servers to Windows Server 2008 and desktops to Windows 7.

Replace 3 Scanners with Epson Expression 10000XLs.

GRIN-Global:

The GRIN-Global project is a joint partnership between USDA-ARS NPGS, Global Crop Diversity Trust and Bioversity International. The goal of the project is to rewrite the current GRIN Germplasm Management System in such a way that it can be deployed on any size computer with a minimum amount of effort. The new Germplasm Management System (dubbed GRIN-Global) will support five different

languages, three database systems and install on a single desktop computer. In 2009 the NCRPIS team attended a Technical Steering Group meeting in Ames to discuss and review the achievements of the project in 2009. The highlights of the GRIN-Global team achievements during 2009 are as follows: Developed GRIN-Global client interface (Curator Tool), enhanced the database schema, migrated the current GRIN Oracle database to Microsoft SQL Server database platforms, and designed/developed/tested the search engine and web services (middle tier) for the GRIN-Global system.

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

Seed Research:

Seed research for 2009 included the following genera:

- *Calendula* – Documenting the progress of seed after-ripening at 4 and 23 °C. The primary goal is to document loss of seed dormancy during cold storage. Results are summarized as the experiment progresses.
- *Echinochloa* – Investigating the effects of light and darkness on release of seed dormancy. Accessions of *Echinochloa* seeds expressed all three types of photoblastic response for germination: light-requiring, dark-requiring, and neutral. These results will be submitted for publication to Seed Science and Technology.
- *Helianthus*– Experimented with 9 treatments on 4 accessions of wild sunflower accessions to determine if a treatment can replace the 6-week,[†] cold-stratification treatment normally used to break seed dormancy. Treatments included acidic and basic solutions, GA₃, Ethephon, GA₃ + Ethephon, running water for 7 days, and a cold treatment (7 days at same temperature as running water). The Ethephon + GA₃ promoted germination best on all four accessions. This improvement ranged from minor (10 percentage points better than the next best treatment, which was either GA₃ alone or the 7-day cold treatment) to fairly significant (36 percentage points than the next best treatment, GA₃).
- *Zea* – Provided GRIN data retrieval for Mark Widrlechner and Allan Trapp (ISU graduate student in statistics) to help develop a statistical model to predict the trajectory of seed deterioration over time that can be used to efficiently schedule viability tests in the future.

[†] If accessions for regeneration are chosen further in advance, even longer cold-stratification periods are used.

Germination Testing:

During 2009, Maria Erickson and her crew conducted germinations on over 2,500 accessions.

Curator Group	Major Crop Tested	Total Accessions Tested
Brenner	Amaranth (461)	630
Marek	Flax (215)	461
Millard	Maize populations (476)	616
Qu	Medicinals (62)	62
Reitsma	<i>Cucumis melo</i> (590)	720
Widrlechner	Ornamentals (59)	63
Total:		2,552

Computer Application Development and Graphics Support:

In 2009, David continued his work on computer application forms and reports to retrieve data from the GRIN Database, and used statistical software packages to run descriptive statistics. He continued to serve station needs by providing AutoCAD drawings and large-format printing for professional posters and local use.

Internet website related:

In 2009, David enhanced the station's website (www.ars.usda.gov/mwa/ames/ncrpis) and the Ames Area Civil Rights Advisory Committee (AACRAC) websites by:

- Adding new 'slide viewer' for featured news on station homepage.
- Creating new Video and Book Library web pages for AACRAC.
- Creating a germination test methods area.
 - What was learned from the AACRAC website development is being applied to the station's website and new features that are scheduled for development in 2010.
- Posting of posters, PowerPoint presentations, updating germination test methods, personnel biographic data, and other updates to the station's website as needed.
- Update biographical information on the station's staff.

The following table summarizes the major uses of our website, as determined by the number of times pages are viewed.

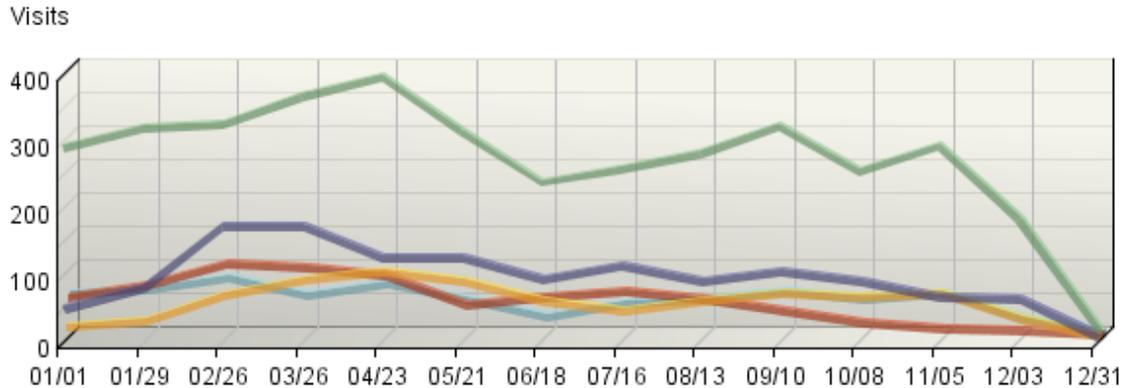
Visit Summary 2009	
Visits	15,635
Average per Day	42
Average Visit Duration (minutes)	4:40
Median Visit Duration (minutes)	1:35
International Visits	7.2%
Visits of Unknown Origin	53.3%
Domestic Visits	39.5%

**Page Viewing Trends for Year 2009
(All Categories)**



In 2009, 36,825 page views were reported. With an average of 100 views per day and 2.35 pages per visit, we are averaging about 42 visitors per day. The marked decrease in December was due to the typical decline in use of USDA websites during the holidays and periods when the WebTrends software that tracks visits was not functional.

**Page Viewing Trends for Year 2009
(Top Five Categories – color key shown in following table)**



Pages Visited (listed here if at least 200 visits)	Number Visits
■ Home page	3,810
■ Research Project: Proposal to Develop New Plant Hardiness Map Data for the United States	1,268
■ News and Events	880
■ Hardiness Zone Maps – China	717
■ Station Information – Ordering Germplasm	705
NCRPIS Staff List	695
Station Information – Crops Maintained	594
People and Places – People at this location	469
Maps	474
Station Information – Print all pages in section	442
Hardiness Zone Maps – Ukraine	405
Products and Services, Publications at this Location	376
Hardiness Zone Maps – Print all pages in section	374
Pollinators at PI – Print all pages in section	368
About Us (Mission Statement)	322
Personnel Page, Mark Widrlechner	321
Maps, NC United States Moisture Balance	306
Germination Test Methods	290
Station Organization Chart	288
Station Information, Station Facts and Purpose	280
Pollinators at PI	265
Personnel Page, Mark Millard	225
Pollinators at PI, Honey Bees	225
Careers, Student Hourly Employment	213
Germplasm Enhancement of Maize	208
Contact Us	208
Research Programs and Projects at this Location	207
Personnel Page, Laura Marek	206
Station Information, Seed Storage Conditions	205
Pollinators at PI, Osmia Bees	202

In 2009, many visitors to our station website were interested in the proposal to develop a new plant hardiness zone map for the US, along with the existing hardiness-related maps that we have posted. The “News and Events” page continues to receive considerable traffic. We need to continue to give periodic attention to this area in order to ensure that it is timely.

The Ames Civil Rights website (www.ars.usda.gov//Main/site_main.htm?docid=8446) was regularly updated with information provided by the NCRPIS Civil Rights Committee representative, Robert Stebbins. This included regular observance information, creating graphics and posting graphics provided by the NCAH graphics department to support observances, changes in personnel, and the posting of committee meeting minutes in PDF format.

Cooperative efforts:

In 2009, David contributed to the following station-wide projects:

- Statistics
 - Station Statistics – these are reported in each year’s annual report.
 - Modified Statistics Programs – adapted statistics programs to meet individual curator requests for data on specific groups of accessions.
- Special printing requests.
 - Brochures
 - Posters
 - Large spreadsheets
 - AutoCAD drawings

Special Training:

Maria Erickson participated in the Association of Official Seed Analysts - Society of Commercial Seed Technologists (AOSA-SCST) Annual Meeting from June 1-6 in Ft. Collins, CO., all business and rules overview meetings, and the voting session.

Plans for 2010:

Results of the *Echinochloa* experiments will be submitted for publication to Seed Science and Technology. Work on *Calendula* will continue, as this is a long-term project to document loss of seed dormancy during cold storage. Research on *Actaea* (dormancy breaking), *Daucus* (enhancing embryo development), and *Zea mays* (optimizing the germination of low-quality samples) will be conducted as time permits.

David hopes to complete the initial deployment of the NPGS Ash Conservation Project website in 2010. In addition, he will work on developing reports for printing labels, inventory reports, and station statistics using a full-featured version of Crystal Reports (to be purchased in 2010).

David and Maria continue to address safety needs in their work environments by ensuring employees under their supervision and coworkers are aware of typical hazards and special procedures and situations that impact their safety.

D. Information Management-Germplasm Collections (R. Stebbins, R. Beyer)**Acquisition:**

The North Central Regional Plant Introduction Station (NCRPIS) acquired 709 new accessions in 2009, the sixth highest amount in the last 14 years. Of these new accessions, 521 were received from within the National Plant Germplasm System (NPGS) through exploration and transfer. This included 157 accessions of ornamentals (half of these were from collection trips conducted by NCRPIS personnel), 91 accessions of *Daucus* from Phil Simon and David Spooner’s collection trip in Tunisia, and 54 accessions of wild *Helianthus* from a collection trip conducted by NCRPIS personnel.

The remaining 188 accessions, received from outside the NPGS, included 89 accessions of ornamentals from various sources and 53 accessions of *Zea mays* subsp. *mays* from the Germplasm Enhancement of Maize (GEM) project.

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

Maintenance:

Robert Stebbins provided curatorial assistance by processing requests for taxonomic re-identifications and nominations of accessions to the inactive file. In total, 75 accessions received taxonomic re-identifications. Among these were 15 accessions of *Chenopodium* and 9 accessions of ornamental crops. Also, 215 accessions were nominated for inactivation, including 78 accessions of miscellaneous crucifers, 47 accessions of wild *Linum*, and 31 accessions of *Helianthus*. Ten of these were inactivated due to duplication. The inventory lots of these accessions were combined with the lots of their respective duplicates.

Additionally, 95 accessions were assigned PI numbers. Included in this group were 29 accessions of wild *Zea*, 27 accessions of ornamental crops, and 20 accessions of *Zea mays* subsp. *mays*.

Projects:

Robert Stebbins worked with Mark Widrlechner to prepare 33 accessions of mints and 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, Robert has returned to entering old passport information from logbooks for early Ames-numbered accessions. During this project, he has encountered duplicate accessions and missing GRIN records, which were then corrected; 327 accessions were reviewed/corrected in 2009.

Robert developed a new form for requesting the assignment of PI numbers to NCRPIS accessions and the instructions for using the form. Robert worked with Lisa Burke and Mark to draft a protocol to ensure that the transfer/inactivation of accessions is properly recorded in the inventory area of GRIN. He also finished drafting instructions for the creation of cooperator records in GRIN and updated order processing instructions.

Robert continues to serve a two-year term on the AACRAC. The primary role of the committee is to promote general awareness of civil rights issues and to foster opportunities for the career development of minorities in agriculture. The committee holds monthly meetings and organizes activities to coincide with nationally recognized observances. Robert works with David Kovach to update and maintain the AACRAC website. We decided to upgrade the list of AACRAC videos available for checkout to include pictures of the video cases, including written summaries of each video's content. Hopefully, this upgrade will better enable

employees to identify and check out videos by interest area. Rachael Beyer scanned the video cases and made the electronic images available.

In October, Robert began a three-year term on the Midwest Area Equal Opportunity Advisory Committee. The committee is sponsored by the Area Director. Committee members play three roles: 1) providing location perspectives to the committee on outreach, partnerships, special-emphasis programs, etc., 2) serving as a location liaison to/from the Area Office to help initiate/coordinate outreach related items at the location, and 3) being a location contact and resource.

Robert also constructed a large poster display stand for the NCRPIS exhibit at VEISHEA, an annual public spring event at Iowa State University. Several staff members volunteered to work at the public display.

Rachael inventoried 2,323 journals and new library acquisitions prior to their inclusion in our holdings and NCRPIS database, thus allowing curators and staff the increased ability to access our collections.

Conclusions:

Compared to 2008, new accessions received at NCRPIS were up by 194 in 2009. Among the maintenance areas, re-identifications were down by 31%, nominations to the inactive file were up by 336%, PI-number assignments were 86% lower, and resolved duplications were 400% higher than the previous year. The number of new accessions acquired and the totals for the four accession maintenance areas noted above were all below the 14-year average.

Plans for 2010:

Robert will continue to enter old passport information from logbooks for early Ames-numbered accessions. He plans to integrate the use of enhanced order action codes into the standard operating procedures of processing germplasm requests. As a result, he should be able to develop and document a method to track shipment costs.

E. Order processing (R. Stebbins, R. Beyer)

During 2009, 2,043 orders were entered into GRIN, a new record for the NCRPIS. These orders led to the external distribution of 26,904 items (primarily seed packets, but also vegetative samples) (Table 3A). Of these, 21,464 items (80%) were distributed within the United States, and 5,440 (20%) were sent to foreign requestors. Additionally, 7,874 items (Table 3B) were distributed within the NCRPIS, for such uses as regeneration, evaluation, and germination and disease testing.

The number of orders entered into GRIN in 2009 was 17% greater than that of 2008; however, the number of items distributed was down by 2,398 or 6%. The number of requests received electronically this year was 1,659, an increase of 21% from 2008.

In order to increase the depth of staff expertise and capacity to handle seed distributions, Candice Gardner requested that Robert mentor Rachael in all aspects

of order processing, including the public GRIN request system, GRIN order-processing functions, methods used to communicate with the curators and plant pathologist, and methods for US Mail and FedEx shipping, internal filing, and the processing of international requests. All germplasm requests that involve only accessions curated by Kathy Reitsma have been processed by Rachael since April, 2009, and Rachael is included along with Robert in all email exchanges regarding pending requests. Since then, Rachael implemented use of the USPS for residential shipments instead of FedEx Home Delivery. This change has reduced our shipping costs to residential customers.

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 area. L. Pfiffner continued to serve as the federal supervisor for several of the crews led by state employees.

In 2009, we stored 2242 inventory lots, including 1024 original seed lots. Of the original lots stored, 449 were *Fraxinus*, along with 100 *Zea*, 83 *Helianthus*, 76 *Chenopodium*, 23 *Daucus*, and 16 *Gymnocladus*. Of the increase lots, 880 Ames increases, 244 non-Ames increases, and 94 check lots were stored. During storage, 127 lots were bulked with previously regenerated samples to create 68 new bulked lots, 65 of which became available for distribution. Of all stored lots, 794 lots were made available for distribution. We split 106 original lots to make them available for distribution in limited quantities. We reviewed 3141 inventory lots for seed quantity, and any discrepancies were corrected in the GRIN database. 836 samples were prepared and transferred to a -20C freezer for long-term storage.

In 2009, 72 accessions received taxonomic re-identification. We re-labeled the 196 seed samples affected and filed the pertinent documentation. In addition, seed samples of 195 inactivated accessions were removed from the active collection and placed in inactive storage. We also re-labeled 155 seed lots associated with the assignment of 95 new PI numbers, and then corrected their cold-storage locations in GRIN to reflect changes in numbering.

We filled 1531 seed orders in 2009, including those for distribution, observation, germination, transfer and backup. There were 480 lots sent to the National Center for Genetic Resources Preservation (NCGRP) for backup, involving both accessions new to NCGRP and supplemental lots for previously supplied accessions. NCRPIS distributed 30,993 packets (the majority filled by seed storage personnel) to meet distribution and observation requests. We transferred 52 inventory lots to other NPGS sites. 21 germination orders were filled which involved 743 lots.

2009 saw the continuation of the prepacking program. With the aid of our student worker, we prepacked 31,092 packets of 1497 inventory lots. A large portion of the prepacking program focused on recently acquired, expired PVP maize accessions. Most of these accessions are in high demand as soon as they are received. Prior to regeneration, the distribution lots (original seed) are prepacked in 15-seed packets. Once the accession has been successfully regenerated, standard distribution

amounts are prepacked from the new lot. In 2009, we received 39 expired maize PVP accessions, which in turn were distributed as 930 order items in 157 orders.

NCRPIS continued to participate in sending seed to the Svalbard Global Seed Vault in 2009, by preparing 1545 accessions for backup there. Each sitecrop was reviewed for newer, good-quality distribution lots, and 7 orders were created. Sample amounts ranged from 200 to 800 seeds depending on the amount of seed needed for two regenerations. For tracking purposes, an inventory action code (SVALBARD) was added to all lots shipped. Packets were filled and orders were sent to NCGRP for repackaging and consolidated shipment.

Seed storage personnel continued to maintain the germplasm distribution display in the farm headquarters hallway. New maps were printed at the start of 2009, and domestic and international distribution destinations were noted.

Scanning of original seed samples continues. In 2009, 604 scans were taken, mostly of original samples. 50 of those accessions were of *Fraxinus* from North American collections. We continue working with Jeff Carstens to streamline the imaging and storing process for *Fraxinus*.

Some imaged samples were new to the station while others were being pulled for regeneration when the entire sample was needed. Creating a visual reference of seed lots that have been used up for planting is important tool to allow future comparisons with the increase lots by curators and storage personnel.

In the summer of 2009, the station continued to participate in the National Science Foundation-funded outreach to Native Americans on Plant Germplasm and Genomes intern program. Two students from the Navajo Nation worked in seed storage to learn the basics of genebank management.

Lisa Burke continues to participate in the development of GRIN Global and attended the GRIN Global Technical Steering Group meeting in Ames in August, 2009.

Lisa Pfiffner continues to work on her studies in Purity Analysis as the primary staff member working with new original seed samples, thus familiarizing herself with many types of seeds and potential contaminants

Lisa Burke became the station CPR/AED/First Aid instructor. After completing a two-day course in instruction, she taught 11 classes in First Aid and 5 in CPR/AED training during 2009. 79 staff members (including 5 from units outside of the NCRPIS) were certified in First Aid and 21 were certified in CPR/AED.

VII. CURATORIAL AND SCIENTIFIC TEAM REPORTS

A. Controlled Insect Pollination Service Program (S. Hanlin, S. McClurg)

Progress:

Caged pollination:

Bee pollinators (minus the alfalfa leafcutting bee) were supplied to 404 cages for controlled pollination of 328 accessions.

Honey bee pollination (Hanlin):

Honey bees were used to pollinate 283 accessions in the field. Honey bees pollinated for Sunflower-Oilseeds: 89 accessions of *Helianthus* and 12 accessions of *Cuphea*; for Vegetable: 60 accessions of *Cucumis*, 24 accessions of *Daucus*, 14 accessions of *Cucurbita*; for Horticulture and Medicinals: 14 accessions of *Rhus*, nine accessions of *Cornus*, four accessions of *Aronia*, three accessions of *Physocarpus*, two accessions of *Spiraea*, four accessions of miscellaneous ornamentals, 14 accessions of *Echinacea*, 11 accessions of *Prunella*, seven accessions of *Hypericum*, two accessions of miscellaneous medicinal plants; for Miscellaneous Umbels: four accessions of *Petroselinum*, three accessions of *Coriandrum*, three accessions of *Orlaya*, two accessions of *Angelica*, and two other miscellaneous umbels.

66 parent colonies of honey bees, 121 double-story nucleus colonies and 11 single story nucleus colonies were overwintered in the indoor wintering facility in late 2008. There was a survival rate of 76% for the parent colonies and 38% for the nucleus colonies, which was lower than last year's 84% and 41%. We left 23 three-story parent colonies outside at two locations and 24 strong three-story hives at three outside locations. All outside colonies were wrapped in groups of two or three with 40 lb. tar paper; the survival rate was 83%. In December 2009, we overwintered indoors 58 parent colonies and 53 double-story nucleus hives.

To prepare for early spring 2009 cage pollinations, we purchased 50 "Buckfast" queens. From our over-wintered bees we made 50 nucleus hives with two frames of brood and three frames of adhering bees in each nuc. We then placed a caged queen in each of those nucs; by mid-May we had productive nucs to place into cages. This protocol allows us to have a limited supply of hives prior to the start of our own queen grafting to fill our early spring pollination requests.

We also supplemented our over-wintered bees with 20 "Buckfast" 3-pound bee packages purchased in mid-April. The packages were placed into full size hives in late spring 2009. Throughout the summer, we collected three local swarms; these were made into colonies and were included in the resources for producing nucleus hives.

In mid-May we selected queens from resilient over-wintered parent colonies to use for producing our own queens for nucleus hives during summer 2009. These queens were set up in cell builder colonies. Queen rearing throughout the summer 2009 produced an average of 32 queens per week. During much of June and July, only

one frame of queens was grafted weekly because of reduced student labor availability. Nucleus hives were produced until early August; hives not used in cages for pollination were fed and strengthened for over-wintering.

In early August, 30 strong double story nucleus hives were made into colonies and all strong single story nucleus hives were doubled to prepare them for over-wintering. Because of low early-fall temperatures and a heavy snow in early December which buried most of the nucs and colonies, the bees in the double-story nucleus hives prepared for over-wintering declined by 60%; no single-story nuc bees survived. The total reduction in colonies was 30%.

As part of regular bee-health assessment, in April 2009, a fourth of the surviving over-wintered hives were sampled for mites. The population range of mites found was 0 to 35 with an average of 15 mites per 100 bees. Because the hives had not been treated for mites in the past two years and mite counts were higher than in the past, both colonies and nucleus hives were given two treatments of Apiguard® (thymol - an organic product) in May. Mite populations in the spring 2009 were determined using the “sugar roll” method: 1 tablespoon of powdered sugar was placed in a pint jar with 100 bees randomly sampled from the hive. Bees were rolled in the sugar and then shaken vigorously over a white pan; mites dropped off into the pan and were counted. As a fall mite control, in September colonies and nucs to be over-wintered were treated with either Apistan^R [fluvalinate] or Check-Mite^R [coumaphos] strips for six weeks. No mite population assessment was made prior to fall treatment, as the spring assessment justified additional fall treatment.

All parent colonies and nucleus hives were given two treatments of Fumagilin – B® in March 2009, after removing them from the over-wintering room. In September 2009, all hives being prepared for over-wintering were given two medicated feedings prior to placement in the over-wintering room. Fumagilin-B® is used for prevention of dysentery [nosema] while the bees are in the over-wintering room.

For wax moth control during the summer of 2009, stacks of supers containing empty frames were treated with Para-moth® (para-dichlorobenzene) crystals on a bimonthly basis to fumigate for moth larvae. Because of the cooler outdoor temperatures during the winter of 2009/2010, the “environmental control method” was used to our advantage. Equipment was stacked in an unheated storage area which was opened to the outside below-freezing temperatures. Wax moth does not tolerate extended periods of extreme cold, so pest emergence was reduced. Because of health concerns with the use of Para-moth® (a carcinogen), we will research more natural methods of wax moth control in the winter of 2009/2010 to determine appropriate methods to use in the stored supers of frames during the summer months.

Use of our syrup feeding system of two 1000 gallon polypropylene tanks (one inside the shop and one outside), a 30 gallon poly “mixing” tank and a dish washer for cleaning syrup jars continues with good success. An immersible heater was used to liquefy the contents of the supplementary storage barrels prior to transfer of that syrup into one of the poly tanks. To prevent crystallizing of the high fructose corn

syrup in the large interior storage tank, the contents were circulated by running the pump for at least five minutes daily.

All bee locations were registered with the Iowa Department of Agriculture and Land Stewardship (IDALS) to assist pesticide applicators in locating bee-yards and notifying appropriate beekeepers prior to spraying nearby. All locations were registered with IDALS using township/range and latitude/longitude. The Lat/Longs were obtained using a handheld GPS unit and each location's coordinates confirmed using "GoogleEarth."

Bombus pollination (Hanlin):

Fourteen "mini-research" colonies of *Bombus impatiens* were ordered during the spring/summer of 2009 from a commercial supplier. *Bombus* colonies were used to pollinate twenty-four field cages with twenty-two accessions. 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 pollen contamination. *Bombus* pollinated for Horticulture: seven accessions of *Monarda*, five accessions of *Caragana*, two accessions of *Spiraea*, five accessions of miscellaneous ornamentals/medicinals; for Vegetable: two accessions of *Cucurbita*; and for Sunflower-Oilseeds one accession of *Helianthus*.

We continued to use 60-quart protective plastic containers to house the cardboard *Bombus* hives while in field cages. The plastic container and hive were placed on a stand consisting of a honey bee hive body and lid; two water-filled quart containers were placed on either side of the hive to prevent the wind from blowing the container and hive off of the stand.

Osmia cornifrons/O. lignaria pollination (Hanlin):

Osmia were used to pollinate a total of 34 field cages and five greenhouse cages with 23 accessions all together. *Osmia* bees pollinated for Brassica-Oilseeds: 20 accessions of *Brassica* (17 in the field and 3 in the greenhouse), one accession of *Crambe* (in greenhouse), and one accession of *Linum* (in greenhouse); and for Horticulture: one accession of *Alcea* (in greenhouse).

We need ca. 1500 *Osmia* bees total each year to supply field cages and to place at "increase" sites for obtaining a supply of bees for the next year's pollination season. We had only ca. 900 bees to use for the 2009 pollination season due to destruction of 2008 increase domiciles from flooding. There were also fewer bees available for purchase from commercial suppliers in 2009.

The 900 bees on hand were used to fill 87 domiciles in 2009; only 39 of these *Osmia* domiciles were used in germplasm increase cages in 2009. Forty-eight domiciles were placed at a single location (The Berry Patch, Nevada, IA) for "increase" bees (to use in the 2010 pollination season). We collected approximately 216 bees from our own increase domiciles in 2009 for use in the spring of 2010. Because of reduced NCRPIS *Osmia* increases in 2009, approximately 1,500 bees will need to be purchased in 2010.

We accurately tracked the physical location of the *Osmia* domiciles at increase sites for retrieval later in the summer (when they may be hidden by foliage), by using a GPS unit and paper maps. As domiciles were placed in the early spring, coordinates were recorded from the GPS unit and each domicile position was plotted on a map. Because this was the first time we used The Berry Patch as our main *Osmia* increase site, the map used to track domiciles was hand drawn at the time of domicile placement; corresponding notes were recorded giving the tree row and the exact tree each domicile was placed in.

Alfalfa leafcutting bee (ALC) *Megachile rotundata* (McClurg):

ALC bees were purchased as larvae in leaf cells from a single supplier for use in 2009. The bee cells were held in refrigerated storage until moved in sets to warm incubation and bee emergence boxes. Bees were available on a weekly basis throughout the year and were used in plant regeneration cages in the field and greenhouse from 7 January through 22 December 2009.

In 2009, 871 ALC deliveries were made. A total of 11 fields and greenhouses with 150 cages containing 159 accessions representing 19 genera and 62 plant species/subspecies received ALC bee pollinators. An additional 15 deliveries were made to 2 *Daucus* accessions with 2010 increase lotcodes in Nov-Dec 2009. These are not included in the 2009 delivery totals, but they are included in the seasonal cage counts in the table below.

Numbers of active ALC cages (those cages where ALC bees have been supplied at least once) ranged from an overall low of 2 cages/accessions in November and December to an overall high of 67 cages with 64 accessions in June. The average number of active cages differed by season as did the number of active cages that were re-supplied with bees weekly. In the winter there are fewer ALC cages but most active cages need to have fresh bees added weekly, primarily because the bees are being incubated outside of their normal range of use. In the summer there are larger numbers of active ALC cages but fewer cages need to receive new bees weekly. The frequency of re-supplying ALC bees in the summer is determined by the attractiveness to the bee of both the accessions present in the cages and the physical structure and location of the cage. Following is a seasonal summary of low, high, and average number of total active cages/accessions vs. those cages which were actually re-supplied with ALC bees on average.

2009 Season – ALC BEES	Winter	Spring	Summer	Fall
LO no. of active cages/accessions	2/2	16/21	29/28	4/4
HI no. of active cages/accessions	18/18	44/53	67/64	34/33
AVG no. of active cages/accessions	9/9	28/36	43/43	15/13
AVG no. cages supplied weekly	76 %	77 %	51 %	70 %

Breakdown of ALC bee use by project from the smallest to largest users: Horticulture had 10 deliveries to 3 locations with 5 cages/accessions of 4 genera (*Alcea*, *Potentilla* (2 species), *Sorbus*, *Symphoricarpos*) from Apr-Sep; Sunflower-

Oilseeds received 17 deliveries to one location with 4 cages/accessions of *Euphorbia lagascae* from Aug-Oct; 422 deliveries were made to the Vegetable project in 4 locations with 53 cages containing 50 accessions of two genera (*Cucumis* (13 species/subspecies) and *Daucus* (8 species/subspecies) from Jan-Dec; Brassica-Oilseeds also received 422 deliveries to 5 locations with 88 cages containing 100 accessions of 12 genera (*Alliaria*, *Aurinia*, *Brassica* (7 species/subspecies), *Crambe* (2 species), *Erysimum* (8 species), *Isatis* (3 species/subspecies), *Lepidium*, *Linum* (3 species), *Matthiola* (3 species), *Sinapis*, *Thlaspi*) from Mar-Oct; of the deliveries to Brassica, 35 cages/accessions were *Camelina* (3 species) in graduate student Ivan Ayala's summer field plot.

In 2008, ALC were tried along with flies for the first time in *Euphorbia* cages. ALC did not compete well with the flies; we speculate that this was due to the limited plant material available in the cages. In 2009, ALC bees again were placed in *Euphorbia* cages along with flies, as larger plant populations were present. Limited ALC activity was noted in some of these cages. While it does appear that ALCs do pollinate this genus, whether adding ALC actually increases seed production over flies alone is unknown.

We made few changes in ALC handling protocols during 2009. We received a single 18 gallon shipment of the bee cells from one supplier as opposed to one small winter shipment and one large spring shipment from two different suppliers as done previously. Amounts of bee cells placed in the incubation containers for summer were adjusted to allow for more equal distribution between weekly bee emergence boxes.

Flies (Blue Bottle Flies and Houseflies) (McClurg):

Fly pupae of two species (Calliphoridae and *Musca domestica*) were purchased from two suppliers and incubated for use weekly from 16 January through 3 November 2009 for caged plant pollinations in both the greenhouse and field. In 2009, 1218 fly deliveries were made. A total of 11 fields and greenhouses with 142 cages containing 153 accessions representing 25 genera and 55 plant species/subspecies received fly pollinators.

The overall average number of cages/accessions supplied with flies in 2009 was 24 cages/ 27 accessions per week with a high of 70 cages/accessions in mid-June and a low of 1 to 3 cages/accessions in January and December. The low to high and average number of cages/accessions supplied with flies by season is shown in the table below. Flies were used during 47 weeks of the year with none needed in early January or the month of November.

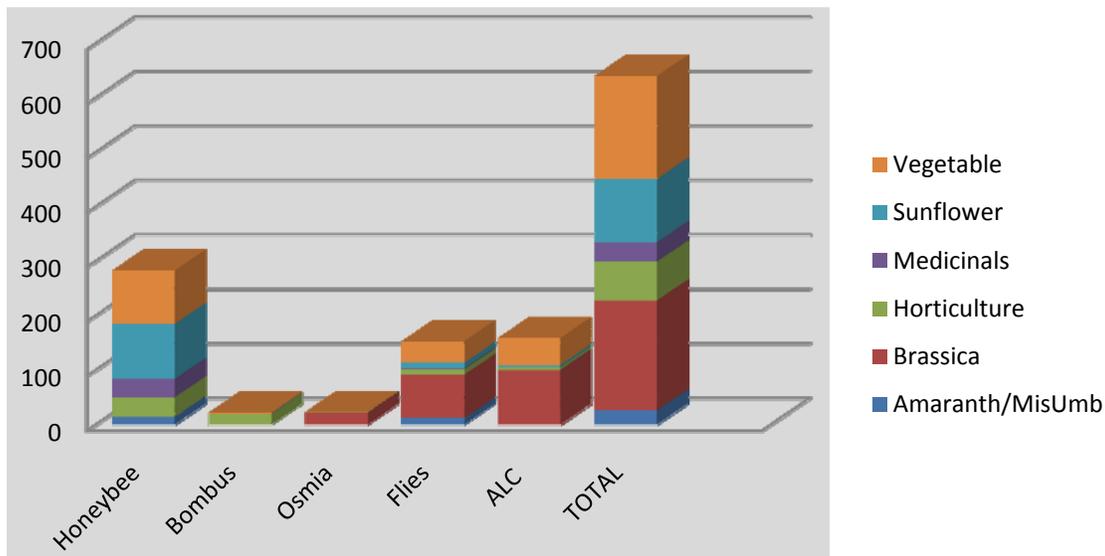
2009 Season - FLIES	Winter	Spring	Summer	Fall
LO number of cages/accessions	1/1	10/14	28/36	1/3
HI number of cages/accessions	9/12	30/44	70/70	26/28
AVG number of cages/accessions	4/5	22/33	50/50	16/18

Breakdown of fly use by project from the smallest to largest users: Medicinals had 13 deliveries to 2 locations in 2 cages/accessions of *Hypericum perforatum* from Jul-Sep; Horticulture had 35 deliveries to 2 locations with 10 cages/accessions of 4 genera (*Ampelopsis*, *Alcea*, *Rhus*, *Spiraea*) from Apr-Sep; Miscellaneous Umbels obtained 62 deliveries to 2 locations with 15 cages containing 12 accessions of 5 genera (*Angelica* (2 species), *Bifora testiculata*, *Coriandrum*, *Orlaya daucoides*, *Petroselinum*) from Jun-Aug; Sunflower-Oilseeds received 129 deliveries to one field with 10 cages/accessions of *Euphorbia lagascae* from Jul-Oct; Vegetable received 451 deliveries to 2 locations with 41 cages containing 39 accessions of 10 species/subspecies of *Daucus* from Jan-Oct; Brassica-Oilseeds received 437 deliveries to 4 locations with 29 cages containing 45 accessions of 12 genera (*Alliaria*, *Aurinia*, *Brassica* (5 species/subspecies), *Crambe* (2 species), *Erysimum* (8 species), *Isatis* (3 species/ subspecies), *Lepidium*, *Linum* (2 species), *Matthiola* (3 species/subspecies), *Sinapis*, *Thlaspi*) from Feb-Nov; in addition to these Brassica maintenance increases, 91 deliveries were made to one field with 35 cages/accessions of 3 species of *Camelina* in June for graduate student Ivan Ayala's study.

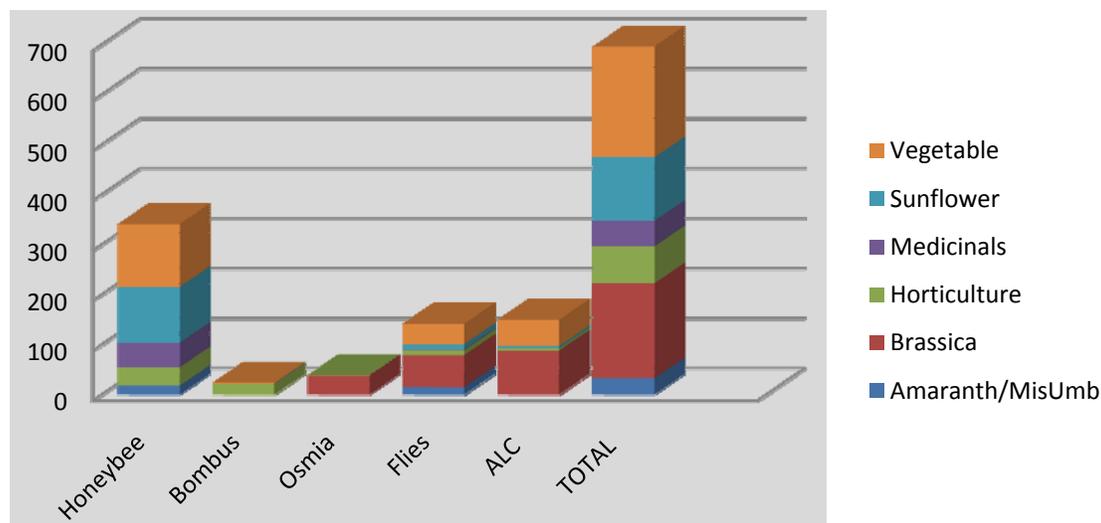
There were few changes in fly incubation or distribution protocols in 2009: only blue bottle fly pupae were distributed weekly in winter and spring greenhouse cages due to few cages of umbels present; both blue bottle flies and houseflies were distributed weekly to summer field cages. Re-supplying flies weekly to cages ensures continued presence of fly pollinators; if appropriate and available, bee pollinators may be present in the same cages.

<u>Summary of Pollinators supplied to regeneration cages in 2009</u>						
Number of Unique ACCESSIONS per pollinator						
	Honeybee	Bombus	Osmia	Flies	ALC	TOTAL
Amaranth/MisUmb	14	0	0	12	0	26
Brassica	0	0	22	80	100	202
Horticulture	36	19	1	10	5	71
Medicinals	34	0	0	2	0	36
Sunflower	101	1	0	10	4	116
Vegetable	98	2	0	39	50	189
OVERALL	283	22	23	153	159	640
Number of TOTAL CAGES per pollinator						
	Honeybee	Bombus	Osmia	Flies	ALC	TOTAL
Amaranth/MisUmb	18	0	0	15	0	33
Brassica	0	0	38	64	88	190
Horticulture	37	21	1	10	5	74
Medicinals	49	0	0	2	0	51
Sunflower	112	1	0	10	4	127
Vegetable	125	2	0	41	53	221
OVERALL	341	24	39	142	150	696

2009 Accessions per Insect Pollinator



2009 Cages per Insect Pollinator



Pollinator Request database and Pocket Pollinator (McClurg):

No changes were made in the current version (3.4) of Pocket Pollinator in 2009. Since most permanent curatorial staff are now familiar with this program version, there were fewer “user errors” this year. McClurg did work with two NCRPIS staff members and a graduate student new to Pocket Pollinator in spring/summer 2009, teaching them the basics of the PDA program and helping them set up Query-embedded Excel workbooks to track their requests as well. Other emphasis was on continued improvement/efficiency of entomologist’s record keeping and improving

written protocols. Copies of “Pocket Pollinator Protocols” were posted on the Entomology area of the NCRPIS SharePoint site for easy staff access.

Tests (Hanlin/McClurg):

Comparison of high fructose corn syrup (HFCS) and baker’s fondant sugar as nucleus hive feed for honey bee:

To continue a 2008 late-summer study in which we compared the use of two food sources (HFCS and fondant) by nucleus hives, two more comprehensive studies were set up in the summer of 2009. These studies compared the use of fondant sugar (recommended by visiting scientists in 2007) and HFCS which has been used as a standard feed by many beekeepers. The studies were designed to: 1) determine if nucleus hives both in and out of caged environments would continue to produce brood and stay strong and healthy, and 2) determine if feeding fondant would be less labor intensive than feeding HFCS.

For the part of the first study looking at free flying beehives only, we chose 12 double nucleus hives. These were set up as three groups of four and within each group, two hives were fed HFCS and two were fed fondant; each of the three groups began their test feeding at different periods during the summer. The first set of four nucs began feeding in May, the second set of four in June and the final set in July. It should be noted that the nucs in this study were required to gather their own food from nearby flowers before they started test feeding.

From the beginning of May, all 12 nucs were checked every two weeks and if swarm cells were present, the research nuc was split to keep all test nucs equal strength. We wished to track the strength of the 12 original research nucs through the summer; this was indicated by the total number of nucs made from each research nuc and included the number of frames of bees and brood removed from each. Once feeding was started on a new group of research nucs, the sets were checked weekly. Data collected included: total frames of honey/nectar, total frames of brood, and total frames of bees.

It was found that the free-flying nucleus hives showed highly significant differences in the amount of brood produced during the summer. Less brood was produced in early summer and increased brood was produced throughout the rest of the summer. The difference in brood production was not related to the type of feed (HFCS or fondant), however. The amount of feed used by the research nucs differed by date but not by the type of feed (HFCS or fondant). There was an increase of feed used both early and late in the summer; the amount of feed consumed by date groups was significantly different while the type of feed was not significantly different. In conclusion, free-flying nucleus hives’ overall strength was not found to be statistically different based on the type of feed they received during the summer.

Again for the first part of the study but looking at caged research nucs only, we used a total of 30 single story nucleus hives with three different genera (*Daucus*, *Cucumis*, and *Helianthus*) with the cooperation of two curatorial projects (Vegetable and Sunflower-Oilseeds). In each of the three crops, we placed five research nucs fed fondant and five research nucs fed HFCS. The research nucs were started as pairs (i.e. a new fondant nuc and a new syrup nuc were started at the same time), as

new pollinator requests were received for the specified genera during the summer. The nucs were removed when a pollinator ‘out’ request was received with only the requested research nuc being removed but not necessarily the original partner started at the same time; i.e. all research nucs in the study ended at different times during the summer.

Hive strength (based on frames of bees and brood) was found to be significantly different among all 10 of the research nucs tested in each of the three genera. This may be due to unequal nuc strength at the start of the study; also, hive strength was reduced when nucs lost their queens or became “drone layers” during the summer. The two food types were supplied to the research nucs during separate work sessions; however, there was no significant difference in the number of times that fondant and HFCS were fed.

The second goal of the summer 2009 study was to determine if feeding fondant would be less labor intensive than feeding HFCS. It took roughly 25 % of the amount of time to feed the research nucs fondant as opposed to the time required to feed with HFCS.

Thus, the overall results for the entire summer 2009 test showed that baker’s fondant sugar could be used as a satisfactory replacement feed for HFCS during the summer in caged nucs used for pollination. It should be noted that free-flying hives are not fed during certain portions of the summer; these hives must receive HFCS early and late in the season to dispense medication and for the hives to prepare adequate food stores. Because of the 2009 results, in 2010, HFCS will be fed during the spring and in late summer, fondant will be used as the primary feed during most of the summer. We will increase the portions of fondant from a 0.25” block used in 2009 to a 1” block in 2010 to further reduce the time spent feeding. Commercial sources of fondant will be contacted in winter 2009/2010 to determine a reliable source of adequate quantities of fondant at a reasonable price.

Compare covered vs. open stands for elevation/protection of honey bee and bumblebee colonies within large *Cucurbita* pollination cages:

In spite of the inconclusive results due to a very wet growing season and lack of vegetation in most of the *Cucurbita* (pumpkin) cages in 2008, we continued to use the covered elevated stands constructed in 2008 in increase cages in cooperation with the Vegetable curators during 2009. A total of 15 pumpkin cages receiving honey bees as pollinators had stands placed at the north and south ends (i.e. two honey bee nucs per cage). Two cages containing *Bombus* as pollinators had a single stand placed at the north end of the cage (i.e. one *Bombus* hive per cage). Based on observations in 2008, the bees appeared to be hindered by the chicken wire fronts on the stands, thus in 2009 we placed the hives so the entrance was opposite the covered fronts.

In spite of the cool wet spring again in 2009, we did observe greater amounts of vegetation in the cages. Vines grew over the stands as anticipated and in some cases fruit formed on the top of the stands. During the growing season, numerous fruit were observed on the outside edges all around the cages. However, few fruit were observed in the dense vegetation in the center of the cages.

In conclusion, it seems that two nucleus honey bee hives placed in raised stands at either end of the large *Cucurbita* (pumpkin) cages or a single *Bombus* hive in a raised stand at the north end of a cage appeared to give adequate fruit production throughout most of the cage. Based on the results in both 2008 and 2009, Vegetable and Entomology staff will continue to use these metal stands to raise insect hives placed in the large *Cucurbita* cages in the future.

Use of multiple ALC domiciles and distributions in *Cucumis* field cages in order to increase fruit production:

ALC bees were used in wild *Cucumis* field cages undergoing first-time regeneration at NCRPIS in 2008. Unfortunately, low numbers of fruit developed in some of these cages, even though ALC bees appeared active to entomologists all season long. In assessing ways to improve fruit production without increased time/ labor investment by curatorial staff (such as use of internal trellises in cages), Vegetable curators questioned the low number of deliveries of ALC bees to some of these cages. Normally, we resupply active ALC cages only when the bee population seems low or no flowers are being visited by bees. The curators speculated that older ALC bees were no longer effective at pollination and wondered if new ALC bees should be introduced every week as is done for fly pollinations.

Vegetable staff observed there was greater fruit formation near the ALC domiciles within the 2008 cages – especially in 5-foot tall cages densely filled with foliage. We know from other cucurbit studies that it is difficult for pollinators to navigate through such foliage-filled cages, and it would be reasonable to assume ALC would visit flowers closer to the domicile in these conditions. Vegetable curators thus wondered if placing multiple ALC domiciles in cages would result in increased fruit production.

We did two informal field studies in 2009 to see if *Cucumis* fruit production in field cages would increase if ALC bees were supplied on a weekly basis and if multiple domiciles were provided. In the first study, three 7-foot tall cages (the 5-foot tall cages are being phased out) were designated to test weekly vs. normal ALC bee deliveries. All three of these cages received multiple ALC domiciles (5 domiciles spaced equally around the cage perimeter) as well, to determine if resulting fruits were nearer domiciles. Two of these cages contained the same accession of *Cucumis dipsaceus*; one of the cages received bees weekly and the other cage received bees per normal (when the bee population was low). The third cage contained an accession of *Cucumis ficifolius*; it received ALC bees every week.

ALC bee domicile activity was observed while bees were being released to these cages during the 2009 pollination season; it was apparent that none of the domicile locations was favored over others by the ALC bees. The two cages containing *Cucumis dipsaceus* received ALC from 26 June to 10 September 2009. Ten deliveries were made to the weekly treated cage, and 5 deliveries were made to the normally treated cage. The weekly treated cage produced 413 fruit while the “normal” cage produced 290 fruit. The cage containing *Cucumis ficifolius* received 7 deliveries of ALC bee weekly from 24 July to 10 September 2009 and produced 338 total fruit. Leaves and vines were plentiful in these three cages at the end of the

field season; it seemed that fruit was located throughout the cage and not related to the locations of the multiple domiciles.

For the second study, which assessed the impact of the position/quantities of ALC domiciles provided in a cage on the location/numbers of fruit formed within, McClurg randomly selected cages from the 2009 *Cucumis* increase plot which appeared to have the most vegetation as of 16 July 2009 when she set up domiciles. Four 7-foot tall and three 5-foot tall cages received multiple domiciles as described in the study above. Another four 7-foot tall cages and one 5-foot tall cage received only single ALC domiciles. All of these cages received ALC bees only on a normal distribution schedule (when bee population was low) due to limited resources.

The conditions for cucurbit field cages were different in 2009 than 2008 in several ways. Most of the *Cucumis melo* grown in the 2009 field increase plot were from a collection undergoing first-time regeneration at NCRPIS; curatorial staff could not predict which accessions might produce excess foliage. We had a cool wet spring/summer in 2009 which resulted in slow growth of foliage. There was increased use of 7-foot tall (7 foot wide) cages in 2009, which greatly increased the amount of surface area available for *Cucumis* vines and thus flower exposure to pollinators, as opposed to the smaller growing area in the 5-foot tall (5 foot wide) cages of 2008. None of the cages selected for the second study in 2009 developed as many leaves/vines as had appeared in some of the 5-foot tall 2008 field cages with poor fruit production.

Cages in the second study received 2 to 4 ALC deliveries from 16 July to 28 August 2009. For 5-foot cages: where multiple domiciles were present fruit numbers equaled 0, 18 and 19, while 2 fruit were formed with a single domicile present. For 7-foot cages: where multiple domiciles were present fruit numbers equaled 2, 6, 7, and 36; no fruit production resulted in the cages with single domiciles. Fruit locations were “mapped” in these cages to assess the impact of domicile placement. Most cages with multiple domiciles had fruit formed throughout the cage. For the one cage with a single domicile where fruit was formed, the fruit was located at the opposite end of the cage from the domicile.

In conclusion, it appeared that weekly bee deliveries and presence of multiple domiciles both resulted in greater *Cucumis* fruit production in the two informal 2009 studies. It is felt that the use of the larger 7-foot cages likely played a part as well. Because the weekly bee distributions occurred only in cages which had multiple domiciles present, we do not know if weekly bee distributions for cages with single domiciles would also result in increased fruit production. If the study is repeated in future on a formal basis, it is suggested to try four treatments: weekly bee distribution with multiple ALC domiciles, weekly bee distribution with single ALC domiciles, normal bee distribution with multiple domiciles and normal bee distribution with single domiciles, in order to determine if one or both practices are equally or greater in importance for cucurbit fruit production.

Further, if weekly bee distributions and multiple ALC domiciles per cage are beneficial for *Cucumis* fruit production, entomologists wonder if this is also true for other crops such as *Brassica* and ornamental accessions that are the most typical

users of ALC bees. This would be wise to test but hard to accomplish at present with reduced resources.

Seed pests – Determine if red and/or gray sunflower seed weevils are present in perennial sunflower field plots and search for better control methods:

The Sunflower-Oilseeds curation team has experienced an ongoing problem with larvae of seed pests destroying critical levels of increase seed in some perennial sunflower plots (especially *Helianthus tuberosus*) at NCRPIS. We felt the most likely pests were red or gray sunflower seed weevils (*Smicronyx fulvus* LeConte and *Smicronyx sordidus* LeConte). The life cycle of both weevils is similar, with adults appearing in the field in July, feeding on sunflower bracts then buds as available. When buds open, adults move to feed on pollen and lay eggs in the developing seeds. Larvae feed within the seeds and when mature drop to the soil in late summer/early fall for overwintering.

Beginning in 2006, McClurg researched integrated management techniques suggested for these pests in commercial cultivated sunflower fields. It was difficult to apply the most effective weevil controls of crop rotation and cultivation to the perennial sunflower plots which must be left relatively undisturbed as long as seed increase is needed from a particular accession. These plots (which are ca. 200 foot square and can have plant populations up to 200 plants) are cultivated only around the perimeter in the spring. Marek and Larsen had implemented use of a trap crop to attempt to draw the weevils away from the increase plots as well as applying at least one chemical spray during the growing season to control the weevils along with other pests. As of early 2009, Larsen felt their current management/spray schedule was not adequate as seed was still being lost.

McClurg researched controls for other ground-dwelling weevils in permanent plantings such as orchards and ornamentals which could potentially be adapted for the perennial sunflower growing conditions at NCRPIS. It may be necessary to attempt to control the overwintering larvae in the soil prior to adult weevil emergence in July, if contact sprays are not an effective control of the adult weevil. Once the adults have deposited their eggs in the sunflower seeds, the eggs and young larvae are protected from any chemical controls and seed loss will result.

One of our goals in 2009 was to determine the exact species of sunflower seed weevil present in the NCRPIS plots as well as the emergence date and population size in order to fine-tune chemical applications to the plot. To trap emerging insects, we utilized wire cone traps designed to monitor populations of nut curculios in pecan orchards (another weevil that overwinters in the soil). It was speculated these traps may serve as a weevil control as well. McClurg monitored the plot for weevils in 2009, rather than Sunflower-Oilseeds personnel as in previous years.

McClurg constructed 6 cone traps using bronze varnished screen (mesh 18×14 .011) cut 3 feet wide and 6 feet long for each trap. Three pieces of 1” diameter PVC pipe ca. 32” long were used to make an internal tripod to support the wire screen. The traps were sewn together with nylon fishing line and PVC tripod legs held in place with fine wire. The top ends of the PVC pipe were plugged with number 2 rubber stoppers to prevent trapped insects from hiding within the legs. The peak of the

wire cone was left open 0.5 inch from the top; a 3" diameter plastic jar lid (for pint jar) with 1.5" center hole was caulked in place below the opening at the peak. The pint jar served as the receptacle for catching insects emerging from the area of soil covered by the wire cone.

The cone traps were placed in the field plot in mid-July 2009; there were few sunflowers blooming in this field at the time of trap placement. Six plots had been flagged by Larsen as potentially "highly-infested" by sunflower seed weevil; three of these plots had already been screened for insect pollination in the 2009 field season. Because of the large size of these plots (cages are 10' × 10' × 20'), McClurg placed two traps within each of the three caged plots. One trap was placed in the north half of the cage and the second trap in the south half. An attempt was made to place traps as close to plants as possible without damaging any of the plants. There was at least one square foot of soil area covered by each cone trap once it was in place.

McClurg surveyed all 26 plots in this field for weevil presence weekly or every-other-week once weevils were discovered in one of the cone traps. Caged plots containing honey bees for pollination were not included in later surveys; it was not feasible to enter the cage safely and cage screens prevented accurate observation of the tiny sunflower seed weevils (2.5 to 3.5 mm) from the outside. The cone traps were not effective in capturing additional sunflower seed weevils through the growing season, although they did serve to pinpoint initial weevil appearance in the field.

The first gray sunflower seed weevil was found in a trap on 22 July; 2 to 40 weevils were counted on plants on the exterior edges of open plots at the same time. Seven plots were sprayed with Malathion to control weevils on 24 July. Only two plots were found to have significant numbers of gray weevils on 27 July (counts of 12 and 24) but 9 plots were sprayed that week to control Japanese Beetles defoliating the plots, as well as sunflower weevils still present after the initial spray. The first red sunflower seed weevil was noted in the field on 29 July; few weevils found in other plots that same date indicated good control by the earlier spray treatment. In mid-August only two plots were found to have higher numbers of weevils (both red and gray species present now); McClurg recommended spraying the entire field at this time due to the presence of a wide variety of insect pests (especially aphids) which were impacting plant health. By the end of August/early September, it is thought that only red sunflower seed weevils were present in the plot and weevil numbers had reduced greatly; isolated cages/plots were hit by more aphids and lepidopteran pests and sprayed for such with Orthene (5 plots on 24 August, 2 plots on 2 September and 1 plot 9 September). By this time in the season most plots were now caged and had honey bee pollinators which complicated weevil surveys.

Most field plots (21 of 26 total) had fewer than 7 weevils throughout the 2009 growing season. Seven plots had higher totals, ranging from 12 to 84 weevils, counted during a 7-week period. McClurg felt there was a greater population of gray sunflower seed weevils present than the red species in the 2009 growing season.

Total seed harvests from the 2008 and 2009 growing seasons were compared to assess the effectiveness of increased monitoring and chemical sprays in 2009. The 2008 plots were sprayed only once in mid-August while there were six spray sessions

in 2009 from 24 July to 9 September, with each plot sprayed at least twice during the 2009 growing season. Overall it appeared less seed was harvested in 2009. Thus it did not appear that increased chemical sprays based on pest populations present were immediately effective in increasing seed harvested from these plots in 2009. However, it is anticipated the 2010 weevil populations should be reduced as a result of the increased spray effort in 2009 and should result in increased seed harvest in 2010.

What else could be attempted to achieve greater seed increase in these perennial sunflower accessions? Perhaps it would be wise to move undamaged tubers to new field plots, which would lack populations of overwintering weevil larvae to attack plants in the next growing season. Another alternate control that McClurg would recommend Sunflower-Oilseeds staff consider, is use of beneficial nematodes applied to the soil of the current infested field plots. These beneficial nematodes are recommended for control of other weevils whose soil-dwelling larvae are known to attack plant roots such as strawberries or ornamental plantings.

Safety:

McClurg and Hanlin thoroughly updated the Entomology building chemical inventory and placed printouts of current inventory on the various storage locations as recommended at an ISU EHS training session. McClurg also compared the inventory data to the Entomology building MSDS notebook. Missing MSDS were replaced with up-to-date versions and the MSDS notebook index was updated as well.

McClurg assisted C. Block and M. Erickson with thorough laboratory inspections of the Plant Introduction chemical laboratories in the Agronomy Building on the ISU campus.

JSA-PPEs created in 2008 were reviewed and “polished” with supervisor Gardner in 2009.

McClurg assisted L. Lockhart in preparing the Kodak Industrex Instant Photo Processor and the Hewlett-Packard Faxitron Cabinet X-ray system respectively for disposal and placement on excess property. The photo processor was considered unsafe for continued use after a leak was discovered. Because the x-ray system was rarely used and digital x-ray images can be obtained from other cooperators, it was decided to remove the equipment from NCRPIS use rather than purchase a new photo processor.

Equipment:

McClurg assisted L. Marek and crew and D. Brenner with the operation of the Virtis “Freezemobile” lyophilizing unit throughout 2009 for drying *Helianthus* and *Amaranthus* specimens. After summer use required adjusted techniques to achieve adequate vacuum with this older unit, McClurg re-wrote the operation instructions.

Presentations, Outreach:

On April 6, Hanlin sent documentation to Thomas Vitrano of the Illinois Federal Correctional Institution on how to rear and introduce the pollinators used at

NCRPIS. This information was requested to promote the pollination of many of the garden products being produced at the facility.

On May 5, S. Hanlin and S. McClurg gave a tour of NCRPIS Entomology facilities to Brindha Narasimhamoorthy and Sue Ruin from Kemin Industries. These representatives were interested in setting up a greenhouse cage program incorporating insect pollinators for the production of rosemary seed. They were provided our *Bombus* hives supplier information and greenhouse cage measurements. Several times in the fall of 2009, S. Hanlin was contacted by Sue regarding both use of *Osmia* and *Bombus*.

On May 14, S. Hanlin spoke at the Squirrel Hollow Outdoor Classroom to approximately 180 sixth graders in 7 sessions about beekeeping and honey bees.

On July 1, S. Hanlin and S. McClurg participated in the farm tour (explaining NCRPIS pollinators) given as part of the RTAC meeting in Ames.

During the August 11-12 – GRIN-Global TSG meeting at NCRPIS, Ames, IA: S. McClurg gave an illustrated talk on Pocket Pollinator. We both participated in the field tour, explaining use of insect pollinators and providing further question time on the use of PDA programming.

In August 2009, we were recognized by the USDA-ARS Midwest area with an Equal Opportunity Award, for assistance we have given to Native American summer interns participating in the Plant Germplasm and Genomics outreach program.

On October 13, S. Hanlin and S. McClurg provided an explanation of NCRPIS pollinators for participants of the Borlaug Fellows Tour.

Several times during 2009, Hanlin was contacted by D. Peterson of the North Central Soil Conservation Research Lab in Morris MN inquiring about native pollinators and *Osmia* bee pollination of *Cuphea*.

Plans for 2010:

Honey Bee Health:

Because of the need to use both types of mite treatment on honey bees in 2009, additional mite population sampling will be conducted in the spring of 2010. This time sugar roll sampling will be combined with the use of a “sticky board.” The “sticky board” method consists of placing a piece of plywood coated in vegetable oil on the bottom board and adding a single treatment of Apiguard® into the middle of the brood frames; after 24 hours all mites that drop and adhere to the sticky board are counted. If 20 or more mites are found, infested hives will be treated with additional Apiguard® in the spring. If no mite treatment (i.e. the additional Apiguard®) is applied in the spring of 2010, hives will again be sampled for mite populations in the fall 2010 using only the sugar roll method. If fall mite populations are 30 or more mites per 100 bees, hives will be treated with Apiguard® at that time. If sampling indicates that mite treatment is needed in both the spring and fall of 2010, Mite-Away® (formic acid) will be used during the fall treatment.

The hives will only be treated once in 2010 with Apiguard® in order to reduce the likelihood of development of resistance of the mites to that product.

Osmia Increases:

To ensure an adequate number of *Osmia* bees in 2010 for cage pollination, three commercial suppliers were contacted in 2009, and 500 bees were requested from each. All suppliers were able to fill their portion of the request, and 1500 bees were obtained to use in both cage pollination and “increase sites” for the spring of 2010.

To prevent flooding losses of *Osmia* domiciles which occurred in the spring of 2008, we will place equivalent numbers of “increase” domiciles at several productive locations rather than a large number of domiciles at the single location at Des Moines Water Works Park as in previous years.

Native pollinators in *Daucus* germplasm observation plot:

Vegetable curators have had a *Daucus* germplasm observation plot at the station for the past several years; in 2009 they mentioned that they have seen an interesting variety of pollinators in this plot. In order to determine if there are populations of beneficial native pollinators on the station, entomologists will make observations of pollinators during the summer of 2010 with the cooperation with the vegetable project. Assorted solitary bee domiciles will be placed throughout the 2010 observation plot of *Daucus* in field W-1. The domiciles used will include the following: alfalfa leafcutting domicile with Styrofoam nest block, *Osmia* domicile filled with standard paper tubes, *Osmia* domicile with bamboo tubes, *Osmia* domicile with smaller paper tubes, and a commercial domicile containing a variety of larger bamboo tubes. The domiciles will be placed in the field when *Daucus* begins blooming and will be monitored as time allows. Observations will be taken at random times of the day and all activity around each of the domiciles will be recorded. It will be noted which type of tubes seem most favorable to native pollinators. Specimens of native pollinators will be collected in order to more accurately identify what types of bees or other pollinators are present.

Non-chemical controls of wax moth in stored combs:

In order to reduce the use of para-dichlorobenzene (Para-moth®) crystals for the control of wax moth in stored combs, several less hazardous methods will be tested throughout the summer of 2010. Starting in the winter of 2009/2010, all sorted frames were stacked in one of two methods. One group of supers was stacked at right angles to each other to increase comb exposure to light and air flow which discourages wax moth movement between supers. The other group was stacked directly on top of each other with a sheet of newspaper placed between each of the supers. The newspaper dividers impede the movement of the wax moth larva to additional boxes of frames and concentrate the damage to only a single box rather than to numerous supers. In several of these straight stacks, a “treatment box” (a super with 2.5” diameter hole drilled in one end and used in the past for the distribution of the Para-moth®) was placed on top of every super of frames prior to the newspaper placement. The treatment boxes in these straight stacks may be opened for light and air exposure to combs; it will also allow for introduction of parasitoids to control wax moth eggs or larvae. *Trichogramma pretiosum* is a moth

egg parasite; we will place approximately 400 wasps (received as pupae in Lepidopteran eggs) into stacks of frames via the “treatment box” holes. The parasitoid will be allowed to fly throughout the stacks and the equipment room.

In the summer of 2010, two types of non-chemical traps will be tested in the equipment room. Trap type one will be the “fermented” trap, a one-gallon jug which contains banana peel, vinegar, water and sugar mixture. This trap will be hung near the stacks of frames; it has been documented to be a very good method of attracting wax moth adults in bee-yards. Trap type two will be a light trap (bug-zapper), possibly some benefit in eliminating the adults; however, most wax moth adults are repelled by light rather than attracted to it. An additional control which has been documented as a deterrent for adult moths is to leave the room lights on. We will use this method in the equipment room for at least eight hours each work day.

During the summer of 2010 and the winter of 2010/2011 frames will be examined to determine if wax moth damage is present. If damage is minimal or lacking, the above non-chemical methods will be implemented as standard practice for wax moth control in the future. If moth damage is present however, more safety precautions will be implemented for continued use of the chemical Para-moth® treatment.

Monitor populations of red/gray sunflower seed weevils in perennial sunflower plots:
The Sunflower-Oilseeds team requested that McClurg monitor the 2010 perennial sunflower plot for sunflower seed weevils again.

Develop a custom laboratory safety survey suitable for the NCRPIS Entomology building:

Following a suggestion gleaned from an ISU EHS laboratory safety course taken in 2009, McClurg plans to develop a “custom” lab safety survey for the Entomology project at NCRPIS. This will ensure that work areas in the Entomology building at the PI farm are being maintained in a safe way as suited to the individual project tasks and facilities.

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

Research Activities:

Maize:

We carried out replicated field trials of 292 maize accessions, mainly inbreds, for Stewart’s wilt resistance. Disease development was excellent following inoculation and gave a good range of resistance reactions in the field. The seven best-performing accessions are listed in the table below along with the two highly-susceptible entries for comparison.

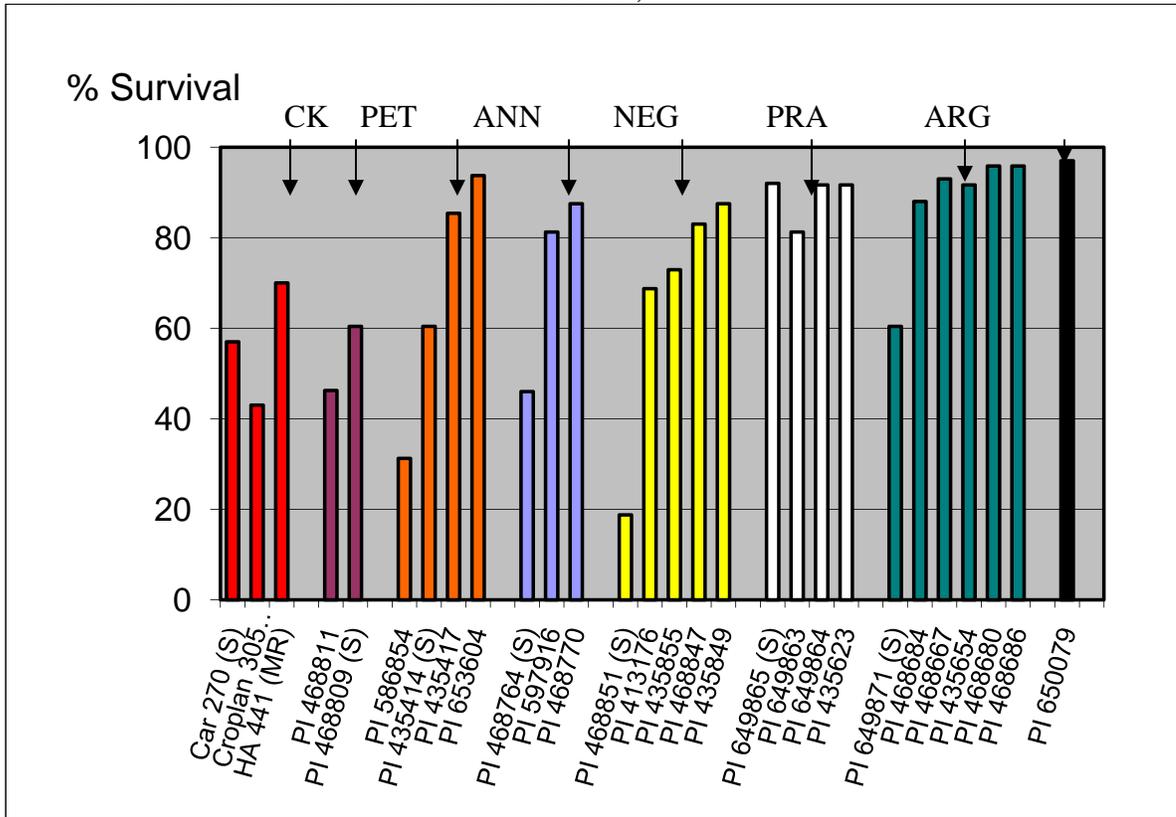
Stewart's wilt resistance of selected inbreds from 2009.

Entry	Alternate ID	Other notes	Average score (1-9 scale of resistant to susceptible)
PI 558533	Mo21R	Resistant to northern corn leaf blight - <i>Exserohilum turcicum</i> . Kernels white, flinty.	1.0
Ames 26754	WXL317	Inbred L317 crossed with waxy gene donor.	1.1
PI 600956	MDF-13D	Yellow corn inbred similar to Mo17. Expired Dekalb PVP.	1.1
PI 601500	PHV63	White corn inbred similar to Pioneer inbred 555. Zapolate Chico in parentage. Expired PVP.	1.2
PI 601569	PHN47	Yellow corn inbred similar to Pioneer inbred 207. Expired PVP.	1.2
PI 601576	PHW79	Yellow corn inbred similar to Pioneer inbred G35. Expired PVP.	1.2
Ames 28932	Mo32W	White corn inbred, Missouri.	1.2
Ames 20140	Mt42	Yellow corn inbred; Minnesota #13 parentage.	6.8
Ames 23507	C42	Yellow corn; Golden Bantam parentage.	8.2

Sunflower:

C. Block and B. Van Roekel conducted evaluations for Sclerotinia stalk rot resistance in wild sunflowers, with co-investigators Dr. T.J. Gulya at Fargo, ND and Dr. L.F. Marek (NCRPIS) at Ames. The work was funded by the USDA Sclerotinia Initiative. In 2009, 247 accessions were screened from species that included *Helianthus exilis*, *H. neglectus*, *H. argophyllus*, *H. praecox*, *H. debilis*, *H. petiolaris*, and *H. niveus*. Greenhouse screening was used to eliminate susceptible germplasm so that only the more resistant material was advanced to field trials. Twenty-six accessions were placed in inoculated field trials at Staples, MN and at Oakes, ND. Entries from each of the species were included in field trials except for *H. exilis* and *H. niveus*. One accession of the resistant perennial, *H. resinosus* was added. Field results correlated well with the greenhouse findings, and many accessions seemed to be more resistant under field conditions. The data from the Oakes, ND trial are shown below. C. Block made oral presentations of Sclerotinia research at the Sclerotinia Research Conference (Bloomington, MN, Jan-2009) and at the National Sunflower Association Summer seminar (Alexandria, MN, Jun-09).

2009 Sclerotinia stalk rot field trials at Oakes, ND.



Disease observations on seed increase crops:

Disease observations were made in the seed increase fields of maize, cucurbits, sunflower, and *Brassica*. Routine disease testing for seed-transmitted squash mosaic virus was also conducted on all cucurbit seedlings prior to transplanting.

Maize:

Field inspections increased substantially (~30%) in 2009 due to the added inspections of the GEM corn plots. The pathology group inspected 3,360 rows of corn and recorded notes for 11 diseases. Many of the diseases were absent and this information is valuable for phytosanitary records. Stewart’s wilt, normally the main disease of phytosanitary concern, was absent as were Goss’s wilt and crazy top. Goss’s wilt was present in the state of Iowa in 2009, the nearest location being 15 miles away. Gray leaf spot and northern leaf blight were present at low levels. Common rust was variable in severity and mainly dependent on susceptibility of the genotype. Seed health tests were run on 234 maize seed lots for various pathogens, the main one being Stewart’s wilt testing on 209 seed lots.

Cucumber species (*Cucumis sativus* and *Cucumis* spp.):

We screened 442 older *Cucumis melo* seed lots for bacterial fruit blotch (BFB) contamination by growing out seedlings under high humidity and temperature conditions in greenhouse isolation and looking for seed transmission. Three infected seed lots were identified. Screening of the entire melon collection (>2300 accessions) is now completed. The cucurbit seed increase plantings (~80 cages) were monitored

regularly for diseases from transplanting to harvest. No BFB was found in the 2009 field plots. The last time BFB was found at Ames was in 2004.

Cucurbit virus-testing:

All cucurbit transplants in the greenhouse were tested by laboratory ELISA for seed-borne squash mosaic virus. Eighty-nine accessions and 2,419 plants were sampled. Two SqMV-infected plants were identified – single plants from two accessions. Results are summarized in the table below.

Squash mosaic virus testing results for 2009.

Species	Accessions tested	Accessions with infected plants	Plants tested	# of SqMV infected plants
<i>Cucumis</i> spp. (<i>melo</i> , <i>sativus</i> , misc.)	73	0	1820	0
<i>Cucurb. pepo</i>	16	2	599	2
Total	89	2	2419	2

Sunflower:

The primary disease of phytosanitary concern in sunflower is downy mildew, caused by *Plasmopara halstedii*. Seeds were treated with the systemic fungicide Allegiance (a.i. metalaxyl) prior to planting. Multiple field inspections were conducted on 600 rows, examining all plants for symptoms of downy mildew, viruses, or aster yellows. The seed increase plants were relatively free from diseases except for *Alternaria helianthi* leaf blight – no downy mildew, virus or aster yellows.

Brassica and related Brassicaceae genera:

The seed increase plots were surveyed for all diseases in late June. Diseases of interest included black rot (*Xanthomonas campestris* pv. *campestris*), blackleg (*Leptosphaeria maculans* or *Phoma lingam*), powdery mildew (*Erysiphe cruciferarum*), *Alternaria* diseases (*Alternaria* spp.), downy mildew (*Peronospora parasitica*) and white rust (*Albugo*). The only two diseases present in 2008 were powdery mildew in several plots and black rot in two plots.

Publications:

Block, C.C., Gulya, T.J. and Marek, L.F. 2009. Evaluation of wild sunflower species for resistance to *Sclerotinia* stalk rot. *Phytopathology* 99:S13.

Safety Topics and Training:

Many hours were devoted to safety topics and training in 2009. C. Block:

- Represented NCRPIS at quarterly USDA campus safety committee meetings and at monthly teleconferences with the Midwest Area safety personnel.
- Wrote, reviewed and updated many JSAs and contributed to the NCRPIS safety committee on JSAs, training and documentation needs.
- Attended EHS classes on: (1) Lab Safety: Laboratory Inspections; (2) Accident Investigation for Supervisors; and (3) Pesticide respirator re-certification.
- Completed online safety training on: (1) Workplace Safety; (2) Hearing Conservation; (3) Epi-Pen Use; and (4) Environmental Management Systems.

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

Acquisition and inactivation:

Sixty-six accessions were acquired (Table 1), including an *Amaranthus*, 9 *Chenopodium*, 3 grasses, a *Marina*, 14 *Melilotus*, 1 *Monolepis*, and 37 Umbelliferae.

Amaranthus pumilus (Ames 30414) is our first accession from Delaware of a Federally threatened species that was already represented by collections from North and South Carolina.

New accessions in the miscellaneous grasses group include *Calamovilfa gigantea* (Ames 29882), and *Glyceria striata* (Ames 29899), donated by the Bureau of Land Management's Seed of Success Program. And the Natural Resources Conservation Service resupplied seeds of PI 518499, an inactivated accession of *Calamovilfa longifolia*, but unfortunately these seeds were inviable upon arrival.

Nine *Chenopodium* were acquired from collections of wild plants: two Great Basin wild species donated by the Bureau of Land Management, five from Brian M. Walsh, a University of Wisconsin graduate student specializing in *Chenopodium*, and two collected by David Brenner.

Marina parryi (Ames 29919) from Arizona was acquired through the Bureau of Land Management.

Melilotus accessions (Ames 30044 to 30056) collected in Russia were donated by Douglas A. Johnson (USDA-ARS, Logan, Utah). Also, *Melilotus* Ames 20475 was separated as seeds from a *Foeniculum* accession from Tunisia.

Twenty-nine of the Umbelliferae are from Tunisia, collected by Drs. Philipp Simon and David Spooner. They include *Carum*, *Cuminum*, and *Foeniculum*. Seven are from wild sources in the United States, donated by the Bureau of Land Management. Two are *Eryngium* from the United States for potential use as ornamentals.

One *Monolepis nuttalliana* (PI 658757) was collected in Boone, Iowa. This is the first accession in our new effort to acquire the wild North American genera allied to *Spinacia*. The accession was featured in a poster about *Spinacia* wild relatives presented at the 2009 American Society for Horticultural Science Annual Conference.



Monolepis nuttalliana (PI 658757)

David Brenner led a collection trip to western Nebraska primarily to collect *Suckleya suckleyana*, but was unsuccessful because the most appropriate habitat was under water in this rainy year. Co-collector Grace Kostel of Black Hills State University will continue to search for the plant during her regular field activities in 2010. We collected soil samples from the area that may contain seeds of *Suckleya* and will attempt to germinate seeds from these soil samples in the greenhouse during 2010. Three accessions (Ames 30075 to Ames 30077) of *Chenopodium*, *Echinacea*, and *Sphaeralcea* were collected during the Nebraska trip.



David Brenner in a Nebraska *Suckleya* habitat.

Maintenance and distribution:

Five-hundred-seventeen accessions were planted in 2009 (Tables 2, 3A, and 3B). Of these, 298 plantings were for regeneration, and 219 were for observations. Most of the observation plantings were grown in single plastic flats in a greenhouse to confirm taxonomy and for other trait observations.

We conducted an experiment on *Petroselinum* (parsley) vernalization. By planting four accessions in March rather than October, with the plants were older and larger at the start of winter. These older plants achieved 100% vernalization, while younger plants of these same accessions had vernalized poorly before. The plants vernalized in our cool greenhouse with night temperatures at ca. 8°C. In our next vernalization experiment, we planted in August 2009, and plan to keep the plants in space-efficient small pots through the winter in a cool greenhouse. In the summer of 2010, the frequency of bolting plants from the August planting will be observed.

There was a fortuitous result of the March parsley planting; in their second year, a plant of one accession (PI 507966) grew unusually tall (7ft 9 3/8 inches, or 237 cm) and was awarded Guinness World Record tallest parsley status. The record was officially witnessed on June 10, 2009 by Jim Popkin of the Ames City Council, and Cynthia Haynes of Iowa State University Extension. ISU and the NCRPIS had positive media attention in newspapers (Heddens, 2009, Majumdar, 2009) and on television. Guinness has the option of including this record in their record book, but they did not guarantee printing.



Our team members from left to right David Brenner, Heather Westemeyer, and Samuel Flomo with the World Record tallest parsley.

Some recently regenerated *Setaria* seed lots have low viability. Of the 68 *Setaria* seed lots regenerated from 2006 to 2009, twelve (18%) had less than 60% viability. Nine (36%) of the 25 *S. sphacelata* seed lots had low viability and none of the 19 *S. italica* lots had low viability. *Setaria sphacelata* is an out-crosser and *S. italica* is a self-pollinator; it is speculated that outcrossing species may be more at risk for this problem than self-pollinating species, but unknown. If the original seed collection has too few self incompatibility alleles, we will be unable to improve on these poor increase lots. However, if improving pollen distribution within the population is helpful, we can possibly improve seed viability by growing larger populations and providing pollinating insects. Since caged flies actively visited the flowers and are associated with high viability seed lots of PI 208949 and PI 307725, we are expanding the use of caged flies in *S. sphacelata* regenerations.

For 10 of the 12 site-crops curated in this project, the number of orders in 2009 was higher than in 2008 (Table 5), so this was an up year for distribution. Many of the orders were from home gardeners.

The number of accessions tested for germination, 630 (7%), in 2009 was higher than in recent years. The numbers fluctuate depending on station priorities, the age of the inventory lots, and normal variations in scheduling.

Echinochloa, Panicum, Setaria, miscellaneous Poaceae:

True-to-type observations on seed lots: We wanted to know if our older, open-pollinated millet seed lots are sufficiently similar to their parental seed lots, due to questions about past regeneration practices; side-by-side plantings were grown for comparison. Twenty-eight accessions of millets were planted in paired 19-ft field rows (in 2008) with the increase seed lots adjacent to the parent seed lot, and, in winter 2009, the same seed lots were planted in single greenhouse flats (1.6ft²). Two accessions (of the 28) that were observed to be dissimilar examples were grown again for a third time, side-by-side in the 2009 field. These field rows can support populations of about 20 plants; the single flats in the greenhouse support populations of about 45 plants.

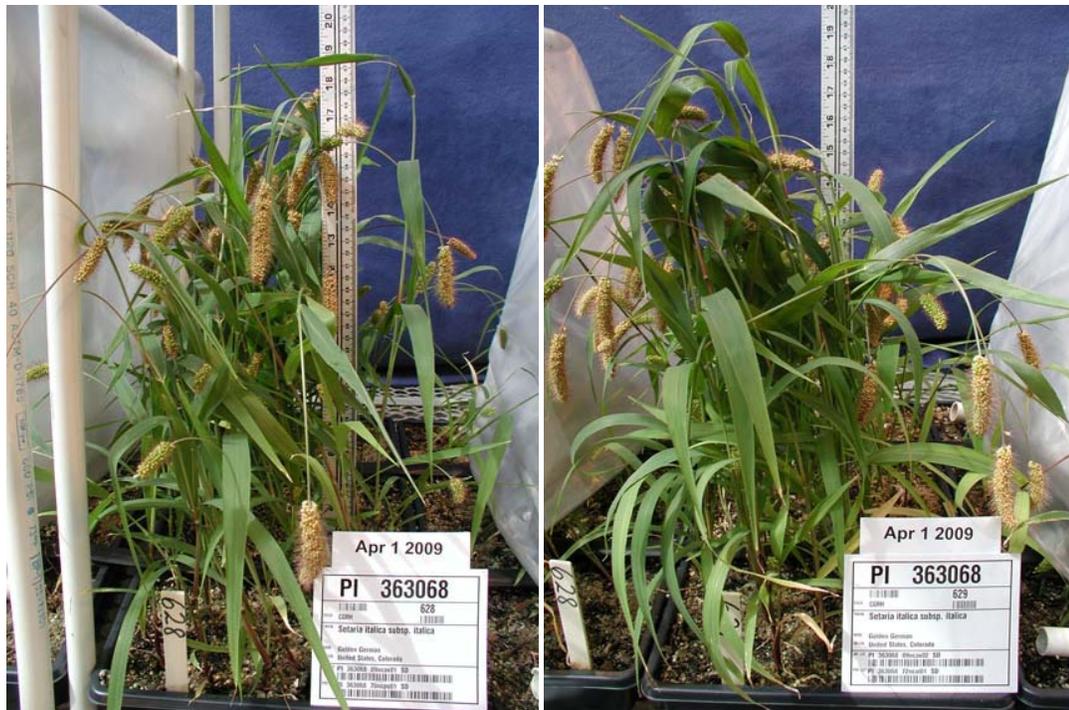
Greenhouse plantings may be superior for this purpose since growing conditions and both con-specific and con-generic weed volunteers can be more controlled. Also greenhouse evaluations require less effort per plant. Greenhouse-grown plants are smaller than field grown plants; 45 plants fit in 1.6 ft² of greenhouse space whereas in the field each plant takes a linear foot of row space. However, the greenhouse plantings are adequate for detecting off-types among these millets such as individuals with different plant coloring, bristle length, and many other traits.

Greenhouse populations of 45 plants per seed lot should detect gross problems in regenerations. If there are questions of contamination by infrequent off-types, the populations should be larger.

In all cases the increased seed lot adequately represented the parent seed lot and demonstrated a heritage of good seed regenerations. This may be partly due to low outcrossing frequencies for our millet species. However, if the study were expanded with more accessions, and replication and/or larger population sizes, we would have a higher probability of identifying dissimilarities between parent-child lots of the accessions with large enough parent seed lots to make such analyses possible.

Observations on selected accessions in the true-to-type evaluation: PI 180304 *Setaria italica* seedlot regenerated in 1988: The original seed lot had some *Echinochloa* which were not in the increase. In the NPGS each accession may only a single species; the *Echinochloa* were properly culled out, and are absent from the increased seed lot.

PI 251273 *Panicum miliaceum* seedlot regenerated in 1959: In the greenhouse two tall, off-types were observed among fifty plants of the increase and none among fifty plants of the parent lot. When it was planted in the 2009 field, two off-types were observed among 28 plants from the increase seeds, but also one off type was observed among 30 plants from the original seeds. I presume that the off type was present originally but increased in frequency during the 1959 increase. I am uncertain whether this accession should be increased again to remove off-types, or not and will consult with the Crop Germplasm Committee. Growing larger sample populations would aid interpretation of infrequent off-types, especially to understand the frequency of the tall off-type in the original seed lot.



Side-by-side plantings of a *Setaria italica* parent seed lot from 1970 on the left and an increase seed lot from 1972 on the right.

Melilotus and other legumes:

Thirty-one *Melilotus* accessions were planted in our cool greenhouse (Farm Greenhouse 2) in October 2009 for spring transplanting into the field. They will be pollinated with caged bees and harvested in mid 2010.

Spinacia and allied genera:

In 2009 seeds of 69 accessions were received from Salinas, California after regeneration there, and 30 accessions were sent from Ames to Salinas for regeneration and harvesting in early 2010. These regenerations are conducted by a cooperative effort between the USDA-ARS and Sakata Seed America, Inc.

An accession of *Monolepis nuttalliana* (PI 658757) was regenerated at the NCRPIS in a greenhouse.

Miscellaneous Umbelliferae:

Our NPGS colleagues in Parlier, California regenerated 19 *Petroselinum* accessions. But they observed only partial vernalization of some accessions and will retain plants for additional flowering in 2010.

We regenerated 17 accessions of seven other Umbelliferae genera in 2009. These include three accessions that were not identified to genus at the time of planting, but were then determined to be *Ammi*, *Oenanthe*, and *Orlaya*.

Dr. Philipp Simon's project (USDA-ARS, Madison, WI) regenerated four accessions, including *Astrodaucus*, *Eryngium*, *Pimpinella*, and *Zizia*.

Characterization/taxonomy/evaluation:

A major data-loading effort with *Melilotus* was concluded, with entry of old data from regenerations that went back to 1990. In 2009, 1,456 *Melilotus* observations were loaded into GRIN (Table 4). The observations involved plant height, days to maturity, flowering time, flower color, and other morphological traits.

In 2009, David Brenner made 46 taxonomic changes in these crops, involving nine different genera, including 14 *Chenopodium*, 8 *Echinochloa*, 7 *Amaranthus*, 5 *Panicum*, and 5 *Orlaya*. Four accessions (Ames 29618 to Ames 29621) donated in October 2008 by Grace Kostel of Black Hills State University as *Monolepis nuttalliana* were grown in 2009 and re-identified as *Chenopodium glaucum* L. var. *salinum*. In addition, 39 existing taxonomic determinations were confirmed and documentation entered in GRIN's Annotation area.

Enhancement and/or utilization:*Amaranthus*:

A study of outcrossing frequency between grain and weedy amaranths under field conditions continues. Samples from ten off-site locations where grain and weedy amaranths occurred together were returned by cooperators to the NCRPIS in 2007 for determination of outcrossing frequencies. Some of these seed lots were grown in a field-demonstration planting, and, as before, the 'Plainsman' (PI 558499) seed lots had more putative crop-weed hybrids (up to 66%) than did the D136-1 (PI 538327) seed lots (up to 4%). In 2010, David Brenner will make a greenhouse planting of these same seed lots to collect data in a more controlled setting.

The same outcrossing study also includes tests with herbicide-resistant weeds. Crosses were made in growth chambers between these same two grain production lines (PI 538327 and PI 558499) and two herbicide-resistant weed accessions (PI 654436 *A. hybridus* and PI 654437 *A. tuberculatus*). F₁ hybrids of these crosses were herbicide resistant as determined by Pursuit® herbicide treatments provided by Micheal Owen and Dean Grossnickle of ISU Agronomy. Five hybrids of PI 538327 × PI 654436 were confirmed per 291 seedlings and 13 hybrids of PI 558499 × PI 654437 were confirmed per 138 seedlings. Herbarium specimens of the hybrid plants were taken for comparison with hybrids from field conditions. Crosses were attempted between PI 538327 × PI 654437 and PI 558499 × PI 654436, but hybrids of those crosses were not recovered.



The red-headed amaranth plants as tall as the identification badge are 'Plainsman', the taller plants are hybrids between Plainsman and weedy types.

Publications and presentations:

Oral Presentations:

Brenner, David M. 2009. The U.S. Spinach Germplasm Collection. T.E. Morelock International Spinach Conference. University of Arkansas, Fayetteville, November 11-12.

David M. Brenner. 2009. Conservation Biology in the National Plant Germplasm System. Guest presentation for the EEOB 531 Conservation Biology Course, Iowa State University. April 30.

Poster Presentation:

David M. Brenner, Grace Kostel, Mark P. Widrlechner, Candice A. Gardner. 2009. Assembling germplasm collections of Nuttall's povertyweed (*Monolepis nuttalliana* (Schant.) Greene) and other spinach (*Spinacia oleracea* L.) allies. HortScience 44:1156-1157. (abstract). Poster presentation at the American Society for Horticultural Science Annual Conference, July 25-28, in St Louis, MO. The same poster was presented at the ISU Agronomy Department Research Symposium Poster session, November 19.

Grant Proposal funded by the National Plant Germplasm System through the Plant Exchange office, National Germplasm Resources Laboratory, USDA-ARS, Beltsville, Maryland:

David M. Brenner and Susan M. Stieve. Plant exploration in Arizona to collect wild relatives of bedding plants and grain amaranth germplasm for crop improvement. To be conducted in September, 2010.

Conferences attended:

American Society for Horticultural Science Annual Conference, St. Louis, MO, July 25-28

T.E. Morelock International Spinach Conference, University of Arkansas, Fayetteville, AR, November 11-12

Crop Germplasm Committee reports:

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

Professional society participation:

In January 2009, David Brenner completed a two-year term as Treasurer of the Association for the Advancement of Industrial Crops.

In August 2009, David Brenner was elected President of the Amaranth Institute by the Board of Directors.

Maintenance:

Passport data:

The International Rice Research Institute collaboration on georeferencing resulted in suggested changes to passport data for about 3,000 of the accessions curated by this project. In many cases, these are new latitude and longitude data from historic place names of collection sites. Processing these changes is a priority, since it should precede new PI number assignment or adding passport information from our paper records. The IRRI data addition results in two versions of GRIN georeference information that should be harmonized into an official version. We must be cautious in handling the data, as IRRI's habitat narrative data will over-write the existing data, and a new PI number may not link-up with IRRI data under a redundant Ames number.

Image processing:

We plan to load into GRIN many of the accession images on file that were taken during seed increases. Samuel Flomo is leading this effort. In 2009, only one image was loaded with 595 images loaded in GRIN over all years (Table 4). There was a delay in 2009 as numerous computer files were re-written to support new image-loading software. In 2010 we intend to load more than 400 images.

Acquisition:

A grant proposal to collect germplasm near Tucson, Arizona was funded for 2010 by the USDA-ARS Plant Exchange Office. This plant exploration is a collaboration between David Brenner and Susan Stieve of the Ornamental Plant Germplasm Center, Columbus, OH. The target area was selected because it is biologically diverse and under-collected for many genera. Collecting there will be aided by the on-line documentation of plant distributions provided by the University of Arizona Herbarium. Collecting targets include species new for the NCRPIS collection in the genera *Amaranthus*, *Gomphrena* and *Sanvitalia*. We will also seek new *Lilium* and *Portulaca* for the Ornamental Plant Germplasm Center.

In the longer term, we need to acquire more species of the bedding plant genera: *Amaranthus*, *Gomphrena*, and *Celosia*, and the spinach allies, *Monolepis*, *Suckleya*, and *Micromonolepis*.

Popular articles about the parsley World's Record:

Heddens, M. 2009. ISU-grown parsley plant breaks record. Iowa State Daily 207(178):3 (June 11, 2009).

Majumdar, N. 2009. World's biggest parsley. The Tribune (Ames, Iowa) 142(1):1 (July 1, 2009).

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

Doust, A.N., E.A. Kellogg, K.M. Devos, and J.L. Bennetzen. 2009. Foxtail millet: A sequence-driven grass model system. Plant Physiol. 149:137-141.

Maughan, P.J., S.M. Yourstone, E.M. Jellen, and J.A. Udall. 2009. SNP Discovery via genomic reduction, barcoding, and 454-pyrosequencing in amaranth. Plant Genome 2:3 260-270.

Trucco, F., T. Tatum, A. L. Rayburn, and P.J. Tranel. 2009. Out of the swamp: Unidirectional hybridization with weedy species may explain the prevalence of *Amaranthus tuberculatus* as a weed New Phytologist 184:819-827.

Upadhyaya H.D., C.L.L. Gowda, V. Gopal Reddy and S. Singh. 2008. Diversity of small millets germplasm in genebank at ICRISAT. 5th International Symposium on New Crops and Uses: Their role in a rapidly changing world, University of Southampton, Southampton, UK, 3-4 September 2007. Pages: 173-185.

Research indirectly related to our germplasm:

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Plans for 2010:

Evaluation:

Taxonomy: In spring and summer 2010, we plan to grow out the *Caucalis* collection (16 accessions) and some reference accessions of *Orlaya* for taxonomic study. The present taxonomic determinations need to be verified or corrected. Drs. Mark Widrlechner (NCRPIS) and David Spooner (USDA-ARS, Madison, WI) will cooperate in this activity.

Melilotus: The evaluation of *Melilotus* is noted as an upcoming milestone on the NCRPIS Project Plan. There is new research interest in winter crops, including *Melilotus*. Our task is to prepare for evaluations to guide decisions on use of *Melilotus* as a winter cover crop. A *Melilotus* poster in 2008 co-authored with Ken Moore was an initial part of the preparation and networking for this effort. In August 2009, 32 accessions of *Melilotus* were planted in 1.5 m (5ft) rows. The plants have established, and their winter-survival will be observed in early 2010. Tom Kaspar (USDA-ARS, Ames, IA) is participating in this effort.

Amaranthus: We are cooperating with the *Amaranthus* cell-wall research of Dr. Olga Zobotina (Iowa State Univ.), who is investigating *Amaranthus* as a bio-fuel crop.

D. Horticulture (M. Widrlechner, J. Carstens)

Acquisition:

During 2009, we worked with Robert Stebbins to enter information for 245 new accessions of ornamentals and mint-family plants into the GRIN database (Table 1). Many of these new acquisitions resulted from the transfer of collections made by the Department of Interior's Seeds of Success (SOS) program in the Western U.S. More than 50 SOS collections of shrubs and herbaceous perennials were transferred from the Western Regional Plant Introduction Station in Pullman, WA to Ames in March. Another major component of the new acquisitions was comprised of *Fraxinus*, most from Illinois, Wisconsin and China. Other important collections included samples of wild populations of *Aronia* (many collected and donated by Mark Brand, Univ. of Connecticut) and of shrubs from southern Wisconsin and northern Illinois collected

during the course of two *Fraxinus* expeditions: an initial summer reconnaissance trip taken by Mark Widrlechner and Jeff Carstens and a fall trip taken by Jeff and Luping Qu.



Jeff Carstens using extendible pole-saw to harvest *Fraxinus* seeds in Wisconsin (left); Mark Widrlechner traversing the backwaters of the Wisconsin River (right).

Plans for the acquisition of *Fraxinus* germplasm from both North America and northeastern Asia moved forward with some urgency in the face of the destruction of native ash populations by the introduction and spread of Emerald Ash Borer. As part of the process to assemble representative *Fraxinus* collections, the USDA-ARS Plant Exchange Office supported reconnaissance and collection trips to Wisconsin and northern Illinois and a collection trip involving Beijing Botanic Garden, to Beijing, Hebei, and Liaoning Provinces in China. In January 2009, Mark Widrlechner chaired a meeting of representatives from the National Plant Germplasm System, the US Forest Service, the Natural Resources Conservation Service, SOS, public gardens, and state agencies with the purpose of developing a coordinated, national collection strategy. This led to the creation of an NPGS collection ash protocol, publicity on about this project through ISU and AP press releases, and the beginnings of an NPGS ash conservation website, which will be released to the public in 2010.

Maintenance:

Maintenance efforts shifted in 2009 towards the regeneration of shrubs in cages, a new field for *Salix* clones, and removal of plants no longer needed after successful regenerations. This was the third year for two cage fields for woody shrubs, one including 42 accessions, focusing on *Cornus*, *Rhus*, *Ligustrum*, *Staphylea*, *Aronia*, and *Physocarpus*, and the other using larger, hoop-house cages for *Cornus alternifolia* and *Rhus typhina*. A new cage field was established for *Aronia* seedling populations received from Mark Brand. Experiments to determine the effects of paclobutrozol in reducing the height and potentially hastening the flowering of *R. typhina* continued and were expanded to include *Fraxinus* seedlings.

The horticulture crew continues to work on mulching long-term field plantings in order to increase plant growth and reduce labor and time requirements associated with mowing. A new permanent planting was established along with side-oats grama-grass (*Bouteloua curtipendula*) to determine the adaptability of a warm-season grass into our production system. All other fields are currently planted with cool-season grasses. Our goal is to reduce the frequency of mowing in permanent planting fields.

Availability:

During 2009, approximately 46% of the ornamental collections and 70% of the mint-family plants were available for distribution (Table 1), figures little changed from those reported in 2008 (45 and 72%).

Regeneration:

Regeneration efforts in 2009 focused on established, caged shrubs, on seed germination for future regeneration cycles, and on the establishment of a new *Aronia* regeneration field primarily for accessions received as seedlings from Mark Brand. The harvests listed in Table 2 include 37 cage increases of shrubs and herbaceous perennials and 17 woody-ornamental seed increases from isolation. There were also 43 accessions of woody plants established from seeds and 12 accessions vegetatively re-propagated. Through these activities and those from the previous year, along with efforts to obtain large, original seed samples, 129 accessions were made available for distribution in 2009.

Viability Testing:

In 2009, seedlots of 55 accessions were tested for germination (Table 2). This included the completion of tests initiated late in 2008, periodic re-tests for stored distribution lots, and new tests on bulked samples. We also conducted cut-tests of more than 430 lots of newly received *Fraxinus* seeds to assess initial quality.

Back-up:

Approximately 40% of the ornamental collections and 74% of the mint-family plants are duplicated at NCGRP (Table 2), figures slightly above those reported in 2008 (38% and 73%). Routine back-ups of dormant *Salix* buds with the National Center for Genetic Resources Preservation in Fort Collins, CO are now underway.



Tests of rooting in cryogenically stored *Salix* cuttings: tests in greenhouse flats (left); rooting evaluation (right).

Distribution:

As summarized below (and in Table 3), requests for accessions of ornamental germplasm increased substantially in 2009, with five-year highs in the number of shipments and the number of different requestors. The 607 “order items” included all plant shipments for the NC7 Trials (described in the following section), along with 16 plants, 140 cuttings, 49 samples for DNA extraction, 8 wood samples, 1 fruit

sample, and 449 seed packets, distributed to fulfill external requests for ornamental plant germplasm. This group encompassed 56 genera; those most in demand were *Calendula* (122 packets), *Fraxinus* (30 packets and 38 samples for DNA extraction), *Cornus* (20 packets, 10 cuttings, and 2 samples for DNA extraction), *Aronia* (20 packets, 10 cuttings, and 1 fruit sample for antioxidant analysis), *Solenostemon* (24 cuttings), and *Tanacetum* (23 packets).

Demand for mint-family germplasm also increased in 2009, with five-year highs in both the number of packets distributed and the number of accessions requested.

Historical Summary of External Distribution Activity:

Crop	Year	No. of Orders	No. of Recipients	No. of Items Distributed	No. of Accessions Distributed
Ornamentals	05	58	53	241	187
	06	89	76	436	322
	07	75	71	268	196
	08	92	83	352	249
	09	110	95	607	390
Mint Family	05	17	16	59	38
	06	19	19	55	37
	07	10	10	54	47
	08	14	14	88	64
	09	33	31	179	92

Characterization/taxonomy:

All the herbaceous ornamentals in the cage fields and many of the tree and shrub accessions being regenerated were checked to verify identifications, and past records were checked as part of the PI-numbering process. In all, 9 ornamental accessions were re-identified. During 2009, Lisa Pfiffner captured seed images of 141 ornamental and mint-family accessions for our local database (Table 4), and Jeff Carstens imaged an additional 12 accessions. These are named following our standard protocol. In 2009, 150 images were loaded to GRIN, by using the mass-loading system for images developed by Pete Cyr.

Evaluation:

Evaluations reported in the 2008 NCRPIS Annual Report are ongoing for two ornamental shrub genera curated in our project: *Aronia* and *Spiraea*. Dr. Mark Brand (Univ. of CT) continues to determine ploidy levels and measure landscape characteristics of *Aronia* as part of a CGC-endorsed project. He is also employing AFLPs to investigate the role that intergeneric hybridization with *Sorbus* may have played in the development of commercial *Aronia* cultivars. And Dr. Mike Mickelbart's laboratory at Purdue Univ. has conducted evaluation of light requirements and pruning regimens on the form and landscape performance of *Spiraea alba* and *tomentosa*, resulting in two manuscripts being prepared for publication.

Enhancement:

There was no major progress to report with enhancement activities in 2009. One small, long-term project to conduct recurrent selection on *Fraxinus ornus* (flowering ash) for improved winter survival continued with the cultivation of a seedling population (Ames 29231) produced from trees selected in Urbana, IL. At the end of 2009, 47 seedlings planted in 2008 at the NCRPIS farm were surviving and will be evaluated for winter injury in Spring 2010.

Coordination of the NC-7 Regional Ornamental Trials:

Plant Distribution - In 2009, Mark Widrlechner and Jeff Carstens distributed 213 plants of five accessions to 16 sites for long-term evaluation, with an additional 74 plants of these accessions provided to 11 public gardens. This year, all accessions were shipped dormant and bare-root.

Web-based planting reports and one- and five-year performance report forms developed by Pete Cyr and Jeff Carstens were used widely by cooperators in 2009 with few problems. These electronic forms drastically reduce the amount of time spent by cooperators and the NCRPIS technician to enter data. Only the ten-year reporting forms remain to be converted to the web-based format.

A brief paragraph with an overview of the NC-7 Regional Ornamental Trials Program has been added to GRIN records for the five accessions distributed in 2009. In addition, links directing Public GRIN users to the NC7 trials webpage have been updated for those accessions.

Germplasm activities in crops other than those curated:

Since 2002, Iowa State University and the University of Iowa have been awarded two grants from the National Institutes of Health (NIH) to establish and support a Center for Research on Botanical Dietary Supplements, which studies health-related effects of *Echinacea*, *Hypericum*, and *Prunella*. Mark Widrlechner continued his involvement with the Center by contributing to the submission of a proposal to renew Center funding and by overseeing a subcontract to ARS, which supports the curation and distribution of the Station's *Echinacea*, *Hypericum*, and *Prunella* germplasm collections so they can be evaluated for chemical composition, genetic diversity, and bioactivity. Details about his involvement and research publications resulting from these efforts are elaborated in the Medicinal Plant section of this Annual Report.

During 2009, Mark Widrlechner was involved with a number of other collaborative germplasm activities including:

1. service with David Kovach (NCRPIS), Dr. Philip Dixon and Allan Trapp, II (ISU) on a project conducted as part of Allan's M.S. thesis to develop and validate a model that predict maize seed longevity from historical, long-term viability test results;
2. collaboration with Kathy Reitsma on taxonomic verification of *Daucus* germplasm, as part of a larger project involving Drs. Philipp Simon and David Spooner (USDA-ARS, Madison, WI), which should ultimately result in the development of a monograph for the genus;

3. collaboration with Kathy Reitsma, Lucinda Clark, and Joseph Kirkbride (USDA-ARS, Washington, DC) to measure pedicel length and elongation in *Cucumis anguria* in an effort to document variation for this characteristic and refine species descriptions; and
4. assisting David Brenner and Susan Stieve (Ornamental Plant Germplasm Center, Columbus, OH) on the development of a plant-exploration proposal to collect germplasm of herbaceous ornamentals and of Amaranthaceae/Chenopodiaceae in Arizona.

Research products:

The development of a reliable protocol for the cryopreservation of dormant vegetative buds in *Fraxinus*, in collaboration with Gayle Volk (NCGRP, Fort Collins, CO), gives us another tool in the effort to conserve North American ash germplasm in the face of Emerald Ash Borer. This protocol allows us to store important clones, especially for staminate and/or sterile cultivars that cannot be conserved as seed populations.

Mark Widrlechner's other research and training activities:

Collaboration continued with George Yatskievych of the Missouri Botanical Garden, which led to the development of draft keys and descriptions for *Rubus* species for an updated "Flora of Missouri."

Collaborations also continued with researchers at Iowa State University and the Chicago Botanic Garden on the development and evaluation of models to predict the risk of naturalization of non-native woody plants. During 2009, we published the results of testing a widely used, risk-assessment model developed by Reichard and Hamilton and three models developed in Iowa on two data sets from the Chicago region in the "Journal of Environmental Horticulture." While the Iowa models were sometimes able to increase classification rates or reduce error rates in comparison to Reichard & Hamilton's original model, no clearly superior model has emerged from the validation process. This work has been expanded to collect and test data sets from Iowa, Minnesota and Missouri through ARS funding for a Specific Cooperative Agreement with Iowa State University, which is supporting Emily Kapler, an M.S. student, who is focusing on regional risk-assessment models. Results from the Chicago effort and a description of our regional plans were prepared in the form of a poster, which Mark Widrlechner presented to the FNRI Researchers' Workshop, Cleveland, OH, in October.

In 2009, Mark Widrlechner continued his service 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 (PHZM) by using the best available technologies and make the next version of the map accessible via the Internet. As part of that service, he assisted ARS personnel in Beltsville in the development of a plan for an external contractor to host the PHZM website and worked with the PRISM group at Oregon State University in preparing a manuscript that describes the technical aspects of map development and the results of that effort. He serves as ADODR on a Specific Cooperative Agreement with Oregon State for that project. Preliminary results from research to develop a model to predict maize seed longevity are quite promising and may lead to the development of software for gene bank

managers. Results were presented to the 2009 Conference on Applied Statistics, Manhattan, KS, in April by Allan Trapp II and an updated version of this talk was given by Widrlechner to the Plant Germplasm Operations Committee, Sturgeon Bay, WI, in July.

Other Horticultural project-training and staff-development activities:

In 2009, Mark Widrlechner and Jeff Carstens attended the Iowa Shade Tree Short Course.

Manuscript and Proposal Review:

Mark Widrlechner continued his service on the Editorial Review Boards of “Genetic Resources and Crop Evolution,” the “Journal of the American Rhododendron Society,” and the “Journal of Environmental Horticulture.” He served as a peer reviewer for manuscripts submitted to three other scientific journals, and as an internal reviewer for two papers prior to journal submission. He also reviewed NPGS Plant Exploration proposals and served as a site reviewer for a proposal by the Reiman Gardens at Iowa State University to receive North American Plant Collection Consortium status from the American Public Gardens Association for their collection of rose cultivars bred by the late Dr. Griffith Buck at Iowa State.

Posters, Presentations and Seminars:

Berhow, Mark A., Brent Tisserat, Sandra Duval, Mark P. Widrlechner, and Candice Gardner. 2009. Nondestructive analysis of phytochemical components by Near Infrared (NIR) spectroscopy: Measurement of rosmarinic acid in *Prunella vulgaris*. Meeting Agenda – Biologically Active Phytochemicals, 49th Annual Phytochemical Society of North America Meeting and Symposia, Towson, MD, 8-12 August. Poster Abstract, p. 37. Available online at <http://www.psna-online.org/PSNA2009abstr.pdf>.

Brenner, David M., Grace Kostel, Mark P. Widrlechner, and Candice A. Gardner. 2009. Assembling germplasm collections of Nuttall's povertyweed [*Monolepis nuttalliana* (Schult.) Greene] and other spinach (*Spinacia oleracea* L.) allies. Poster presented to the 2009 Annual Conference of the American Society for Horticultural Science, St. Louis, MO, 27 July. Abstract published in *HortScience* 44:1156-1157.

Erickson, Maria, Lisa Pfiffner, Lisa Burke, David Kovach, Mark P. Widrlechner, and Candice Gardner. 2009. The NCRPIS – Providing diverse plant genetic resources for worldwide research and development. Poster presented at the AOSA/SCST Annual Meeting in Fort Collins, CO, 3 June. Abstract published in *Seed Technol. Newsl.* 83(2):18.

Kapler, Emily, Mark Widrlechner, and Jan Thompson. 2009. What's going to invade? Regional risk-assessment models for non-native woody plants. Poster presented to the FNRI Researchers' Workshop, Cleveland, OH, 14-15 October.

López, P.A., M.P. Widrlechner, P.W. Simon, S. Rai, T.B. Bailey, and C.A. Gardner. 2009. Applying molecular markers in coriander populations with diverse geographical origins. Presentation to the XIII Congreso Nacional de la Sociedad Mexicana de Ciencias Hortícolas, A.C., Torreón, Coahuila, Mexico, 17-21 August. Abstract published in *Memorias de Resúmenes (Proceedings of Abstracts)*, p. 2.

Trapp II, Allan, Philip Dixon, and Mark Widrlechner. 2009. Predicting the time of 50% seed viability in maize. Presented to the 2009 Conference on Applied Statistics, Manhattan, KS, 20 April, updated version of this talk presented by Widrlechner to the Plant Germplasm Operations Committee, Sturgeon Bay, WI, 14 July.

Publications (other than those involving the Medicinal Plant Collections) which appeared in print in 2009:

Volk, G.M., R. Bonnart, J. Waddell, and M.P. Widrlechner. 2009. Cryopreservation of dormant buds from diverse *Fraxinus* species. *CryoLetters* 30:262-267.

Widrlechner, Mark P. 2009. H. Marc Cathey. *HortScience* 44:220-221. (Reported Deaths)

Widrlechner, Mark P., Janette R. Thompson, Emily J. Kapler, Kristen Kordecki, Philip M. Dixon, and Galen Gates. 2009. A test of four models to predict the risk of naturalization of non-native woody plants in the Chicago Region. *Journal of Environmental Horticulture* 27:241-250.

Widrlechner, Mark P., Kathleen R. Reitsma, Lucinda D. Clark, and Joseph H. Kirkbride, Jr. 2009. Length and rapid elongation of pedicels of the female flowers of *Cucumis anguria* L. *Cucurbit Genetics Cooperative Report* 31/32: 36-40 and back cover .

Departmental Activities:

Mark Widrlechner continued as an active member of the faculty overseeing the Plant Breeding and Genetics major 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 a member of the POS Committee for a Ph.D. candidate in Natural Resources Ecology & Management (NREM) and became a member of POS Committees for an M.S. candidate in NREM and two Ph.D. candidates in Agronomy and in Genetics, respectively. He completed service as a member of a POS Committee for an M.S. candidate in Statistics.

Conclusions and Plans for 2010:

Curation:

Curation efforts in 2009 focused on *Fraxinus* acquisition and seed processing, with samples from more than 300 mother trees processed by the end of the year. Given the serious threat caused by the continued expansion of Emerald Ash Borer in the North Central Region, we will continue to collaborate with Kevin Conrad (National Arboretum), Ned Garvey (Plant Exchange Office), Dave Ellis (NCGRP), Kris Bachtell (Morton Arboretum), and Bob Karrfalt (USDA Forest Service) to refine and execute plans to conserve North American ash (*Fraxinus*) germplasm (and acquire

Chinese germplasm). 2009 was not a very good seed production year for ash in much of the central and eastern U.S., but we are hopeful that 2010 will be more productive. We are planning collection trips to Kansas, Missouri, and northern Arkansas for blue ash, and to southern Minnesota and central Wisconsin for white, green, and black ash for the 2010 field season, and a trip involving the Arnold Arboretum to Pennsylvania and New York in 2011. We will also work with David Ellis to integrate ash seed collections from various sources currently being held in black-box storage, when the passport and seed-quality data warrant and donors permit.

Also related to the acquisition of tree germplasm, we have established a collaboration with Andy Schmitz at the Brenton Arboretum, Dallas Center, IA to assemble collections of *Gymnocladus* (Kentucky Coffee tree), which can be accomplished during the late winter, with plans to obtain samples throughout the south central US early in 2010.

Regenerations in 2010 will focus on producing control-pollinated seeds from the large number of shrub accessions now established in field cages, on unavailable accessions of *Calendula*, in preparation for seed-lipid analyses of the entire *Calendula* collection by Terry Isbell (NCAUR, Peoria, IL), and on germinating seeds for the next cycle of shrubs.

Experiments to determine the effects of paclobutrozol on *Rhus* and *Fraxinus* seedlings will be analyzed in 2010, as part of a broader effort to increase the efficiency of maintaining long-term plantings. For example, in 2010, Jeff will add the use of a pre-emergent herbicide in hopes of reducing time spent on controlling summer annual weeds. Mulching and use of warm-season grasses in long-term plantings will also continue.

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, in 2010, we plan to link them to accession records in GRIN.

With regard to IT advancements, we look forward to the completion and release of an NPGS Ash Conservation website, our testing of new features resulting from the ongoing development of GRIN-Global, and also to the development of a web-based system for the capture and management of all data from the NC-7 Regional Ornamental Trials. The latter will result from GRIN-Global development and implementation.

Ames-numbered, ornamental and mint-family accessions that are currently available for distribution will be considered as candidates for the assignment of PI numbers in 2010, which involves passport-data proofing, identity verification, and duplication checks. Another major passport-data proofing project will involve the review of georeferencing done by IRRRI from historical source and origin records on GRIN. Each set of geographic coordinates and changes to passport data proposed by IRRRI will be checked and, if necessary, corrected before being loaded to GRIN.

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 (for crops other than medicinal plants) will focus on:

1. assembling data on the native ranges and biological characteristics of non-native woody plants cultivated in Minnesota and Missouri and testing risk-assessment models for the invasiveness of a new set of non-native woody plants in Iowa, with an overall goal of developing broader, regional risk-assessment models for the Midwest;
2. completing publications describing technical aspects of the development of the USDA Plant Hardiness Zone Map and horticultural applications of the map along with the PRISM group at Oregon State University and other collaborators;
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; and
4. working with Maria Erickson to develop an M.S. thesis project to investigate factors that may influence differences in long-term viability among maize accessions and with Allan Trapp, David Kovach, and Philip Dixon to identify appropriate viability-testing intervals for stored maize samples that balance the resources used to conduct these tests with the need to identify samples that are declining in viability.

Staff Development:

Plans for staff development for 2010 will focus on training experiences for Jeff Carstens, which are likely to include attendance at the Iowa Shade Tree Short Course, ArcGIS skills, and safety training.

E. Maize Curation (M. Millard, M. Lively, T. Moore)

Equipment:

The maize curation project's main equipment addition, in collaboration with the GEM Project, was an enlarged temporary structure for photoperiod control during the Iowa summer growing season. The structure measured 35' × 102' or 0.8 acre or 0.3 ha and it accommodated 96 – 12.5' long rows. Half was used by GEM to induce flowering on tropical accessions that were used as males for crossing to elite lines. The other half was used to increase tropical maize inbreds and populations that are in high demand and were in low quantity.



Personnel:

The maize curation project full-time staff remained stable in 2009 with Trent Moore (ISU Ag Specialist), Matt Lively (Federal Ag Technician), and Mark Millard (Federal Geneticist/Maize Curator). Federal student labor for maize curation hovers at just under 4 full time equivalents with future reductions expected.

Research Progress:

The maize curator spent a considerable amount of time on the GRIN-Global project in 2009. The project is a collaborative project between The Global Crop Diversity Trust, Bioversity International, and USDA-ARS to create the next generation of the GRIN system. A scalable, database neutral system, it will provide tools and web interfaces for the management of genebank collections and germplasm information, and for their users. The project began in early 2008 and is led by IT Specialist Pete Cyr of Ames. The maize curator, who has a long history of working with the current GRIN system, was assigned to serve as an analyst on the development team. He is tasked with working with GRIN users to identify and provide work flows and system needs to the software developers, and to help develop specifications for the new curatorial software tools. The database project has required over a third of his time in 2009 and is expected to be completed by the end of 2010, when it will be fully available for deployment to international genebanks. Please refer to project details elsewhere in this report and see the project website at http://www.gringlobal.org/index.php/Main_Page for additional information.

Acquisition:

Statistics show a 105 accession increase in 2009 (Table 1) maize holdings over 2008. These include 39 expired Plant Variety Protected (PVP) accessions and 24 GEM releases. There were 6 TZ inbreds of interest that passed through the St. Croix quarantine facility. These lines with resistance to aflatoxin contamination were developed by the International Institute of Tropical Agriculture (IITA) through a collaborative breeding project with Southern Regional Research Center of the USDA-ARS. They show resistance in a kernel-based screening for aflatoxin production and an artificial field inoculation test in Africa to the *Aspergillus flavus* strain found in

Nigeria. These inbreds were sent out to many scientists in the U.S. who will be doing U.S. field screenings during the summer of 2010.

Finally the *Zea* collection saw the first accessions brought into NCRPIS collections under the Standard Material Transfer Agreement (SMTA). The collections are 29 teosinte collections from CIMMYT collected in Mexico. They consisted of 8 collections of *Zea mays* L. subsp. *parviglumis* H. H. Iltis & Doebley and 21 collections of *Zea mays* L. subsp. *mexicana* (Schrad.) H. H. Iltis. Collection quantities ranged from 2,200 cupules to 16,800 cupules. The average for the *mexicana* collections was 5,000 cupules and for the *parviglumis* collections 7,500 cupules. A requestor will need to acknowledge receiving the seed under the SMTA of The International Treaty on Plant Genetic Resources for Food and Agriculture before the seed is shipped. The wording of the SMTA can be found at: http://www.planttreaty.org/smta_en.htm. One request has gone out for this material, giving an initial test to the NPGS GRIN software for handling SMTA requests.

Regeneration:

There were 364 regeneration attempts made in 2009 or 1.8% of the 20,166 accessions on inventory. This compares with 618 (3.1%) of 20,057 accessions in 2008 and 788 (4.0%) of 19,928 in 2007. Again, in 2009 as in 2008 fewer single-row attempts were made than in 2007. Student staffing decreased in 2009 also.

There were 252 accession regeneration attempts made in Ames in the summer of 2009 compared to 377 in 2008 and 588 in 2007. Fewer single-row increases were attempted on original seed in 2009, resulting in fewer accessions being grown, even though the number of rows for increase was approximately equivalent to the 2008 nursery. Increases included 223 inbreds (70 expired PVPs) and 27 populations compared to 160 inbreds (25 expired PVPs) and 217 populations in 2008. There were 87 Australian inbreds increased among the non-PVP inbreds. These included several brown-midrib and waxy lines. Another high priority increase was a group of high methionine conversions of inbreds A632, B73, and Mo17 from the University of Minnesota registered in 2008 in the Journal of Plant Registrations by Philips et al. Only a few populations were grown to reduce labor requirements. The season had a warm dry spell in April and we were able to plant on April 23. It was much cooler than average for the entire summer, making pollination time pleasant, but delayed. We had several very late maturing inbreds in the nursery when a hard freeze occurred in early October. We harvested several of these late lines before black layer maturity and before the hard freeze and dried them slowly at cooler temperatures, avoiding freeze damage and allowing some physiological processes to proceed normally, giving better viability. The adapted inbreds and expired PVPs have very good seed quality. No Stewart's wilt was observed in any increases, which means no ELISA testing is necessary on 2009 Ames increase lots to meet phytosanitary requirements. All factors considered, the 2009 summer maize regeneration year is rated as above average for Ames.

This year's St. Croix quarantine nursery consisted of 116 accessions, compared to 185 accessions in 2008. Included in this nursery were 59 Australian inbred lines, 10 Nigerian inbreds (including reported low aflatoxin producing lines), and the rest were populations mostly from Africa.

Ten tropical inbreds were planted at ICIA in the GEM winter nursery. Since they were later than most of the GEM nursery, a student of Dr. Doebley's volunteered to do the selfing during the time he was working his nursery. Many thanks are given to Mike Blanco and Tony Studer for making this increase possible.

Greenhouse increases included *Zea* accessions consisting of one Gaspé type, three *Zea diploperennis* accessions, and two *Zea perennis* accessions. A *diploperennis* by maize hybrid was also increased. A *Coix* accession was harvested from overwintered plants.

Syngenta received 14 tropical accessions for increase on Kauai, Hawaii. Many thanks are extended to Syngenta for this contribution.

Maintenance:

Table 1 indicates that maize accession availability increased from 65% to 66% at the end of 2009. It also increased by 1% in 2008 and 2007. Effort has been focused on increasing inbreds and expired PVPs to meet demand. Availability reflects this effort although PVP availability lags due to unexpired Utility Patents that restrict distribution although the seed has been increased. In 2009, 377 maize accessions were made available for a net gain of 252 available accessions. This compares with a net gain of 298 in 2008 and 321 in 2007. Flat budgets and staffing make substantially increasing the number of available accessions difficult.

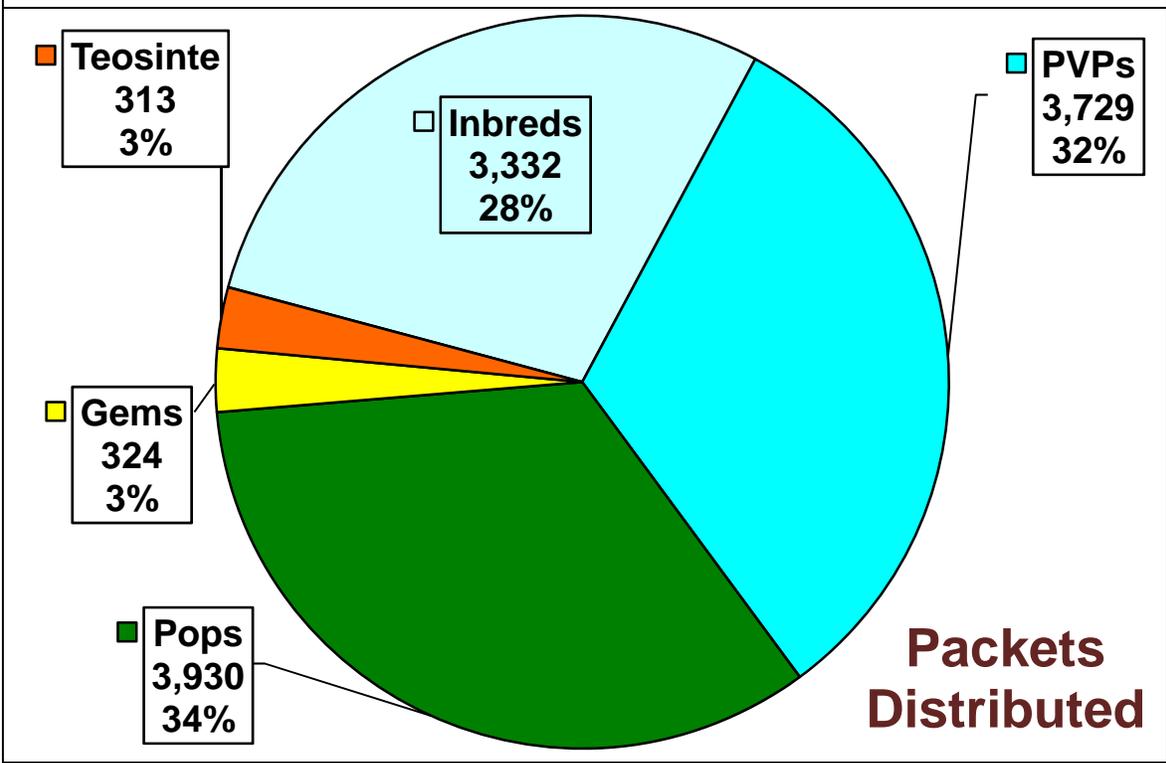
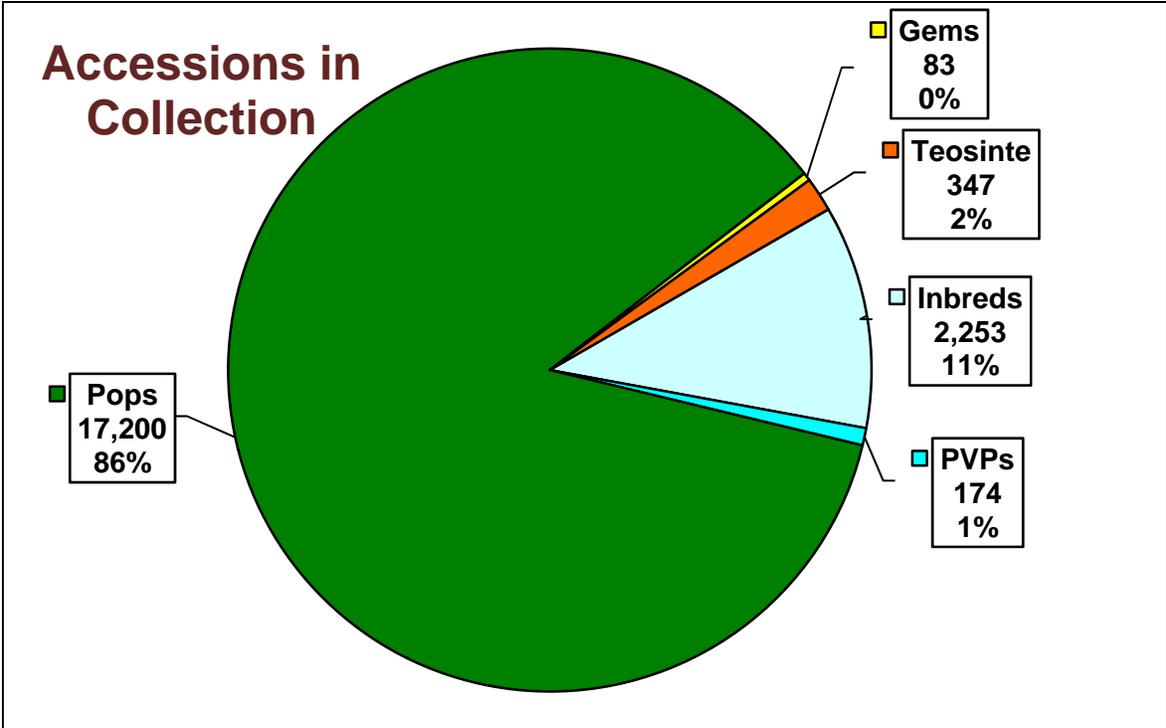
In 2009, 1,209 accessions or 6% of the collection was tested for viability compared to 1,399 (7%) in 2008 and 965 (5%) in 2007. In 2009, 71 accessions were backed up compared to 368 in 2008 and 1,027 in 2007. The percent backup held at 74% in 2009.

Distribution:

Table 5 shows that maize packet distributions (+12%), orders (+28%), and the number of requestors (+29%) were again up significantly over the previous year. This required a much greater amount of time to be devoted by the curator to orders than in previous years. The expired PVP inbred lines continued to be popular distributions. The pie charts below show the contrast in the number of accessions in each group and the number of packets distributed.

Crop	Calendar Year	No. of Orders	No. of Recipients	No. of Items Distributed	No. of Accessions Distributed
Maize	2005	381	275	4425	1828
	2006	585	356	7927	2477
	2007	553	376	8870	2175
	2008	601	406	10474	3457
	2009	768	522	11628	4156
Average		578	387	8655	2819

Zea Subdivisions	Calendar Year	No. of Orders	No. of Recipients	No. of Items Distributed	No. of Accessions Distributed
Populations	2005	149	135	1302	1055
	2006	195	163	2132	1463
	2007	207	174	1722	1016
	2008	234	191	2338	1547
	2009	321	278	3930	2810
Average		221	188	2284	1568
GEMs	2005	13	12	142	66
	2006	28	25	334	66
	2007	23	22	381	67
	2008	27	23	329	81
	2009	25	18	324	87
Average		23	20	302	73
Teosintes	2005	46	43	253	77
	2006	59	49	303	77
	2007	67	62	272	43
	2008	60	58	201	42
	2009	64	55	313	149
Average		59	53	268	78
Inbreds	2005	161	132	1633	604
	2006	265	197	2956	760
	2007	259	203	3314	919
	2008	271	210	5216	1634
	2009	310	237	3332	916
Average		253	196	3290	967
PVPs	2005	103	51	1095	76
	2006	214	95	2202	111
	2007	188	106	3181	130
	2008	190	107	2340	153
	2009	235	124	3729	194
Average		186	97	2509	133



Characterization:

There were 10,242 data points loaded into GRIN on 821 accessions in 2009 compared to 15,641 data points on 2,026 accessions in 2008 and 13,001 points on 2,436 accessions in 2007. Reductions in regenerations and lower student staffing reduced data generation. GRIN-Global duties of the curator reduced the amount of time spent on GRIN loading.

We imaged 599 accessions in 2009 compared to 1,378 accessions in 2008 and 829 accessions in 2007. Again fewer regenerations and lower student staffing reduced image generation.

A nursery of 600 tropical accessions was sent to Monsanto and planted on Oahu, Hawaii. Each accession was planted in a single-row plot mainly to obtain maturity data and adaptation characteristics to determine which accessions could be successfully regenerated in a planned 2010 Monsanto regeneration nursery. Much of this data will be entered into GRIN in 2010.

Evaluation:

Two disease screening nurseries were sent out again in 2009 as in 2008 and 2007. Dr. Bill Dolezal, Pioneer Hi-Bred/DuPont, screened 205 accessions for northern leaf blight resistance and diplodia ear rot screening. Infection was good for both diseases. Lodging and rainy weather in the fall interfered with ear rot results. Dr. Charles Block, USDA-ARS at the NCRPIS, screened 298 accessions for Stewart's wilt resistance.

Communication:

Again in 2009, several tours were given and there were many venues for exchanges of ideas. The GEM project field day continues to generate visits from many maize scientists who are also interested in the maize germplasm collection. The two maize technicians were involved with more of these tours.

Acquisition:

The University of Illinois retired pathologist Dr. Don White has a large collection of inbred lines. We plan to review those holding for important missing accessions and begin receipt of accessions. We are also planning to contact Dr. Zeno Wicks, retired maize geneticist from South Dakota to ensure there are no additional inbred lines or population cycles that are important to add to the maize collection. Dr. James Brewbaker has a collection of tropical inbred lines we will start to incorporate into the collection in 2010.

Fifty-five maize PVPs will expire in calendar year 2010 compared to 35 in 2009 and 40 and 34 projected to expire in 2011 and 2012, respectively. In 2013 there will be a considerable reduction in expirations because most of the certificates granted in 1995 had a 20-year term instead of the earlier 18-year term.

More GEM accessions will be released for distribution by the NCRPIS in 2010.

Regeneration:

Funding will not support a maize-curation project, tropical-maize regeneration nursery in 2010. Efforts will be confined to small increases in the GEM tropical

nursery and voluntary increases by the private sector. Monsanto will be increasing at least 250 tropical accessions on Oahu, Hawaii. A small increase nursery of some 15-30 accessions is expected to be managed by Syngenta on Kauai, Hawaii. Some effort will be put into photoperiod control in Ames. The GEM and Maize Curation groups continue to improve a photoperiod-control system. A challenge is to scale-up the system to needed production levels.

Regenerations in Ames will be maintained at 300-350 accessions annually. St. Croix will be growing 50 accessions of non-quarantine tropical accessions in the fall of 2010 in place of the quarantine increase, because those needing quarantine increase are few.

Maintenance:

Ames-numbered accessions will be reviewed and PI numbers assigned; it is estimated that over 1,200 Ames-numbered accessions and 400 currently available NSL-numbered accessions could be assigned permanent PI numbers. This process is a priority for 2010, following the GRIN-Global project work.

Viability tests will be maintained at 2009 levels. A project analyzing past maize germination results to give a targeted date for the next germination test is underway led by Dr. Mark Widrlechner. This approach should maximize limited viability testing resources.

Evaluation:

Results, of a large 600 accession observation nursery evaluating accessions for a successful increase on Oahu, Hawaii will be incorporated into GRIN.

Dr. Ed Buckler transferred funds to the NCRPIS so that a nursery of ca. 2,500 inbreds could be planted in 2010 and phenotyped at four locations and DNA sampled in Ames for SNP genotyping in Buckler's lab. The phenotyping nursery will be replicated at Raleigh, NC; Columbia, MO; and Ithaca, NY. A federal, term maize technician will be hired to do the majority of the work.

The maize curator will attempt to augment the collection of images currently on GRIN of 5,000 accessions with images of additional accessions in 2010. These images have already been captured, and will be loaded with the new GRIN loader software developed by NCRPIS staff.

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 future use of accessions.

Plans for 2010:

The GRIN-Global project will take a considerable part of the maize curator's time. Regeneration remains this curator's first priority because without viable seed, no other research can be done. Expected reduced student labor staffing due to fiscal constraints will make this a challenge.

F. Medicinal Plants (M. Widrlechner and L. Qu)

In December 2008, Dr. Luping Qu was selected to fill the medicinal-plant curatorial vacancy formerly held by Dr. Joe-Ann McCoy at the NCRPIS, and he began work in March 2009. Since his arrival, he has been learning curatorial procedures and gradually assuming responsibility for the management of these collections from Dr. Widrlechner, who had been curating them until Dr. Qu was hired, with help from Nathan Johnson, the project's STEP employee until July, and Jeff Carstens, technician.

Acquisition:

During 2009, we received and/or collected 40 new accessions of medicinal species, which represents 8% of the current collection (Table 1). The collection currently consists of 489 accessions.

The main focus of germplasm acquisition in 2009 was on *Hypericum* and *Prunella* including native populations that could be obtained in conjunction with efforts to collect *Fraxinus* in southern Wisconsin, northern Illinois, and Ohio. Five new *Prunella* accessions from Asia were also acquired in 2009; three from an ARS-funded fruit exploration to Japan, and one from China and another from Japan, both donated by Quarryhill Botanical Garden (Glen Ellen, CA).

PI Number Assignment:

After proofing passport data and working with Robert Stebbins, we requested and received PI numbers for 13 *Prunella* accessions that were successfully regenerated and otherwise met the requirements for PI-number assignment in 2009.

Availability and Backup:

Sixty-three percent of the NC7 medicinal accessions are currently available (Table 1). In 2009, 21 seedlots of these accessions were made available and 14 accessions were backed up, with a total of 318 accessions now backed up in Fort Collins, representing 65% of the total collection (Table 2).

Regeneration and Maintenance:

Regeneration efforts focused on the completion of caged plantings established in previous years. A new field with 37 new accessions of *Echinacea*, *Hypericum*, and *Prunella* was also established in 2009 for regeneration. Seeds from 30 perennial accessions of *Echinacea*, *Hypericum*, and *Prunella* regenerated in field cages were harvested and processed for storage in 2010.

Viability Testing and Seed Germination Investigation:

Seedlots of 54 accessions were tested for germination in 2009 (Table 2). The testing included recently acquired original samples and those recently regenerated. We also tested seedlots to optimize germination protocols for *Prunella*. The testing conditions included tests in a germinator in light or darkness, direct planting of seeds in flats in the greenhouse, and direct planting of seeds under field conditions. We found that light generally promotes *P. vulgaris* seed germination. Seedling emergence rates in flats and under field conditions resembled those from germinator tests under light. These results will be useful for developing *Prunella* germination and seedling development protocols for regeneration at the PI station.

Distribution:

In 2009, 267 items were distributed; of these, 81% were domestic and 19% were foreign distributions (Table 3A). Along with seed distribution, fresh materials of *Echinacea*, *Hypericum*, and *Prunella* from caged-regeneration plots were sampled by Dr. Ludmila Rizshsky and Mr. Matt Crispin at ISU for metabolomic analyses. In the fall, *Echinacea* roots (9.50 kg dry wt) and *Hypericum* (0.57 kg dry wt) and *Prunella* plants (4.63 kg dry wt) were harvested, dried and ground by Wiley mill and made available to projects associated with the NIH-funded Iowa Center for Research on Botanical Dietary Supplements (Botanical Center) grant project.

Characterization and Taxonomy:

All the medicinal plants in the cage field were checked to verify identifications, and herbarium specimens were taken for accessions that had not been vouchered previously. In 2009, 15 accessions were vouchered and no re-identifications were required.

Digital images of plants were taken for 18 accessions, primarily images of seeds taken by Lisa Pfiffner. Four observation records and 3 digital images accessions were loaded to GRIN in 2009 (Table 4).

Pathogen Observations:

Field plantings were monitored weekly during the growing season for *Colletotrichum gloeosporioides* (anthracnose) and aster yellows disease symptoms. Both pathogens were observed in 2009 on plantings from previous years. A *C. gloeosporioides* seed screening protocol for *Hypericum* continues to be utilized for all newly received and freshly harvested seed accessions. All germination and pathogen data collected have been entered in the GRIN database.

NIH Proposal Development:

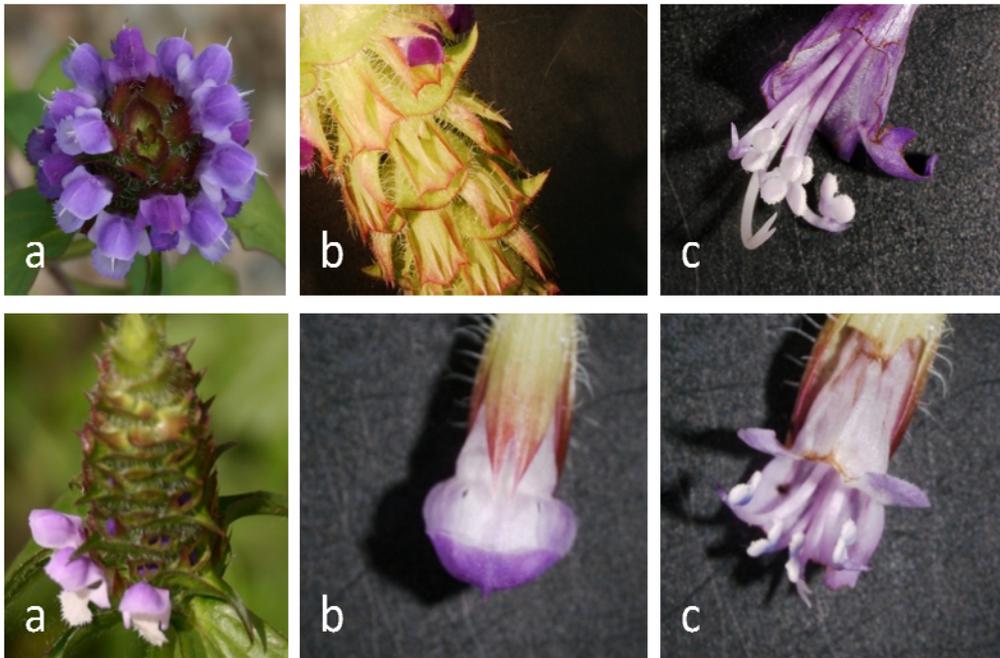
Drs. Widrlechner and Qu actively participated in the development of the Core A (Germplasm and Phytochemical Profiling) portion of a proposal to NIH to renew funding for the Botanical Center, which ends in May 2010. Dr Widrlechner also contributed extensively to editing and finalizing the entire renewal proposal. The proposal was submitted to the NIH in early December 2009.

Analysis of Breeding Systems, Ploidy, and the Role of Hexaploids in Three *Hypericum perforatum* Populations:

The tetraploid cytotype is the predominant type in *Hypericum perforatum*, but hexaploid and diploid plants may also be present in natural populations. Populations with mixed ploidy levels could have detrimental effects on seed regeneration if they can intermate. We have worked with Dr. Shawn Rigby at Iowa State University using flow cytometry to examine the breeding systems of three NCRPIS *H. perforatum* accessions. We have found that hexaploid *H. perforatum* may have minimal effects on seed regeneration in these predominantly tetraploid accessions. A manuscript resulting from this research was prepared and submitted for publication.

***Prunella* Breeding System Investigation:**

We investigated the breeding system of this species in 2009. Two types of floral morphology (pictured below), one with exerted styles extending past open corollas when viewed from above, and the other with shorter, inserted styles, were found among 30 accessions. Two accessions originally collected from Asia uniformly displayed extended styles, and 27 accessions had inserted styles. One accession from Oregon displayed variation in this trait among individual plants. Microscopic observation of 7 accessions, including ones with both exerted and inserted styles, revealed that they all release pollen to some degree before the flowers open (picture below). Using bagged flowers, we found that selfed seed set varied widely among 8 accessions, ranging from 6 to 94%. The accession with 6% selfed seed-set also had extended styles. These findings suggest that mating system in *P. vulgaris* may be in the process of evolutionary change, and that understanding breeding-system variation should be useful in developing efficient seed-regeneration protocols and breeding and selection strategies. Preliminary results from this work, along with those from the flow-cytometry work in *H. perforatum* noted in the previous section, were presented in November by Candice Gardner to the 21st Annual Meeting of the Association for the Advancement of Industrial Crops, Chillán, Chile, and a manuscript resulting from this research is being developed for publication.



Flower morphology types and in-bud pollen release of *Prunella vulgaris*. Ames 29995 (top: a. flowers with exerted styles, b. stage for observation of in-bud pollen release, and c. in-bud pollen release) and PI 656839 (bottom: a. flowers with inserted styles, b. stage for observation of in-bud pollen release and c. in-bud pollen release).

Medicinal Plant Posters & Publications by Mark Widrlechner and Luping Qu for 2009:

Berhow, Mark A., Brent Tisserat, Sandra Duval, Mark P. Widrlechner, and Candice Gardner. 2009. Nondestructive analysis of phytochemical components by Near Infrared (NIR) spectroscopy: Measurement of rosmarinic acid in *Prunella vulgaris*. Meeting Agenda – Biologically Active Phytochemicals, 49th Annual Phytochemical Society of North America Meeting and Symposia, 8-12 August 2009, Towson, MD. Poster Abstract, p. 37. Available online at <http://www.psna-online.org/PSNA2009abstr.pdf>.

Birt, Diane F., Mark P. Widrlechner, Kimberly D.P. Hammer, Matthew L. Hillwig, Jingqiang Wei, George A. Kraus, Patricia A. Murphy, JoeAnn McCoy, Eve S. Wurtele, Jeffrey D. Neighbors, David F. Wiemer, Wendy J. Maury, and Jason P. Price. 2009. *Hypericum* in infection: Identification of anti-viral and anti-inflammatory constituents. *Pharmaceutical Biology* 47: 774-782. Doi: 10.1080/13880200902988645.

Brindley, Melinda A., Mark P. Widrlechner, Joe-Ann McCoy, Patricia Murphy, Cathy Hauck, Ludmila Rizshsky, Basil Nikolau and Wendy Maury. 2009. Inhibition of lentivirus replication by aqueous extracts of *Prunella vulgaris*. *Virology Journal* 6:8 (14 pages). doi: 10.1186/1743-422X-6-8. Available online at <http://www.virologyj.com/content/6/1/8>.

Huang, Nan, Cathy Hauck, Man-Yu Yum, Ludmila Rizshsky, Mark P. Widrlechner, Joe-Ann McCoy, Patricia A. Murphy, Philip M. Dixon, Basil J. Nikolau, and Diane F. Birt. 2009. Rosmarinic acid in *Prunella vulgaris* ethanol extract inhibits Lipopolysaccharide-induced prostaglandin E2 and nitric oxide in RAW 264.7 mouse macrophages. *J. Agric. Food Chem.* 57: 10579-10589. doi: 10.1021/jf9023728

Maury, Wendy, Jason P. Price, Melinda A. Brindley, ChoonSeok Oh, Jeffrey D. Neighbors, David F. Wiemer, Nikolas Wills, Susan Carpenter, Cathy Hauck, Patricia Murphy, Mark P. Widrlechner, Kathleen Delate, Ganesh Kumar, George A. Kraus, Ludmila Rizshsky, and Basil Nikolau. 2009. Identification of light-independent inhibition of human immunodeficiency virus-1 infection through bioguided fractionation of *Hypericum perforatum*. *Virology Journal* 6:101 (12 pages). doi: 10.1186/1743-422X-6-101. Available online at <http://www.virologyj.com/content/6/1/101>.

Qu, L., and M.P. Widrlechner. 2009. Variation in breeding systems in *Hypericum perforatum* and *Prunella vulgaris*. Talk presented by C. Gardner to the 21st Annual Meeting of the Association for the Advancement of Industrial Crops, Chillán Chile. Abstract published in: Cermak, S.C. and M. Berti (eds.) 21st Annual AAIC Meeting – The Next Generation of Industrial Crops, Processes, and Products: Program and Abstracts. Gran Hotel Termas de Chillán, Chillán, Chile, p. 44.

Wu, Lankun, Philip M. Dixon, Basil J. Nikolau, George A. Kraus, Mark P. Widrlechner, and Eve Syrkin Wurtele. 2009. Metabolic profiling of *Echinacea* genotypes and a test of alternative taxonomic treatments. *Planta Medica* 75: 178-183.

Plans for 2010:

Regeneration:

Prunella vulgaris and *Hypericum perforatum* will be the focus in 2010. We will attempt to germinate original seed samples of all accessions of these species that have not yet been successfully regenerated. Based on field observation, these two species could mature seeds in their first growing season, if germination is scheduled early and seedling development enhanced during the initial spring greenhouse growing period. Winter damage of plants of some accessions of these species has been a hindrance to seed regeneration, so we will start germination of the seeds in

January 2010 and transfer the seedlings into larger cell containers to grow larger plants of an advanced development stage for field planting.

Characterization and Evaluation:

We will establish a planting of field plots with 10 or more plants each of at least 10 accessions of *Hypericum perforatum* and *Prunella vulgaris* for data collection following standard descriptors for *Hypericum* and to refine draft descriptors for *Prunella*.

Investigations of breeding systems in *Prunella* will continue on the extent of selfed seed set through in-bud pollination under typical regeneration conditions (caged with bees). The focus will be on the accessions that displayed high seed set when the inflorescences were bagged.

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

Project management:

Due to budget considerations, full-time oilseeds technician, B. Bingaman was released on June 30, 2009. Farm worker, L. Crim was assigned to work on the oilseeds project half-time.

Acquisitions:

We received 118 new oilseed accessions in 2009.

Helianthus:

Eighteen cultivated *Helianthus annuus* accessions, all with expired property rights protection (CSR), were requested and received from NCGRP, Ft Collins. One of the new cultivated accessions was increased in 2009, 14 will be increased in 2010 and the remaining three increased in 2011. Eight accessions of *H. argophyllus* were collected in 2009 by Dr. Sue Thompson in Queensland, Australia. Five accessions for which there were not enough seed to allow immediate distribution will be increased in 2010. During a 13-day NPGS Plant Exchange Office (PEO) funded plant exploration trip to Kansas, Oklahoma, Arkansas and Missouri, Dr. Marek and USDA sunflower botanist Dr. Gerald Seiler, Fargo, ND collected 51 accessions of *Helianthus* from wild populations [*H. annuus* (2), *H. occidentalis* ssp. *plantagineus* (3), *H. pauciflorus* ssp. *pauciflorus* (16), *H. salicifolius* (18), *H. silphioides* (11), and *H. ×laetiflorus* (4)]. Seventy-eight percent of the new wild accessions will be available as original seed when accessioning is complete. The new collections represent the first time germplasm for *H. salicifolius* will be available in amounts large enough to allow for standard distributions. The new collections also contain the first accessions in the NPGS of the Arkansas ecotype of *H. occidentalis* ssp. *plantagineus*. One of the three accessions collected will be available after accessioning is complete. A local collaborator in Kansas provided seeds from an additional five wild populations. Five accessions were received from Shaw Arboretum, Missouri, and three wild sunflower accessions were transferred from the W6 Regional Plant Introduction Station, material wild collected in a cooperative project with the Bureau of Land Management (BLM).

Brassicaceae:

Seven new *Brassica rapa* accessions collected from naturalized populations in southern California were donated by Dr. Cynthia Weinig. One accession of *B. napus* was received from a PEO-sponsored, wild carrot collection trip in Tunisia. In addition, five Brassicaceae accessions were transferred from the W6 Regional Plant Introduction Station, material wild collected in a cooperative project with the BLM.

Linum:

One accession was received from the Shaw Arboretum, Missouri, and four accessions were transferred from the W6 Regional Plant Introduction Station, material wild collected in a cooperative project with the BLM.

Miscellaneous asters:

One accession of *Eutrochium maculatum* was received from a collection trip to Wisconsin by the NCRPIS Ornamental Group. Nine accessions were transferred from the W6 Regional Plant Introduction Station, material wild collected in a cooperative project with the BLM.

Collection Maintenance:

General statistics about availability and management of the collections are presented in Tables 1 and 2 in the appendix. Fewer regenerations than average were started for the field in 2009 for all oilseed groups due to the replacement of a full-time technician by a half-time, inexperienced staff person. Selected details for oilseed accessions increased during 2009 are noted below.

Helianthus, Ames increases:

Cultivated *H. annuus* accessions are 95% available. We are managing our increases to maintain a high level of availability and to ensure that the core collection accessions are available. In 2009, 42 *H. annuus* cultivated accessions were regenerated. Cultivated *H. annuus* accessions requiring long seasons or short days to flower are increased in the NCRPIS greenhouse as space allows. Three accessions were increased during the winter of 2008-2009. Wild annual *Helianthus* accessions are 94% available and wild perennial accessions are 60% available (20% available five years ago). We caged 10 wild annual *Helianthus* accessions and harvested seed from eight of the accessions. Seed was harvested from 49 of 61 caged perennial accessions, 35 of which had been previously established in the field.

Helianthus, Parlier alternate grow-out site:

We continue to work with NPGS Parlier, CA personnel to increase wild taxa that require longer growing seasons than are reliably obtained in Ames. The Parlier environment also provides a valuable alternative for growing mountain and desert species such as *H. exilis* and *H. deserticola* that have not done well in the mid-western humidity and heavy soils. The Parlier location has 40 sunflower cages, purchased by NCRPIS, and can grow up to 40 sunflower accessions per year. In 2005 we implemented the increase protocol of germinating seed in Ames and shipping live seedlings to Parlier. The Parlier staff transplant seedlings and manage plant growth. Accessions are caged before flowering and introduction of pollinator insects; harvested material is shipped to Ames for threshing and processing. In 2009, we sent seedlings for 26 accessions, 25 of which were harvested. In addition, one accession established in 2008 was re-caged and harvested in 2009. The 2009 harvested material arrived in Ames in November and December and is being processed.

The Parlier group records basic field data (date transplanted, dates of harvest) but does not have the staff to record standard descriptor data such as ray and disc flower color, plant height, or branching characteristics nor to take images. Because some accessions represent taxa which we never see growing in Ames, it is important that the observation data be captured. In September 2009, Irv Larsen, oilseeds project technician and Cindy Clark, NCRPIS vegetable project technician, traveled to Parlier to record descriptor information and take images.

Brassicaceae:

Brassicaceae accessions are 88% available. We continue to work towards having at least 90% of this division of the oilseed collection available. In 2009, field populations for 26 Brassicaceae accessions, 24 *Brassica* and 2 miscellaneous crucifers, were established. Seed was harvested from 21 of the field-established accessions. Three accessions did not flower and were transferred to the greenhouse at the end of the season. Two *Brassica* accessions were infected with black rot and the plants destroyed. The accessions will be regenerated in 2010 from treated seed. In addition to the fall-transferred material, 40 Brassicaceae accessions were maintained in the greenhouse, 37 of which flowered and were harvested during 2009. Many wild Brassicaceae in the collection are of Mediterranean origin and could be expected to flower during cool, moist, short-day weather. For example, *Erysimum* accessions have not flowered in the field in Ames but have done relatively well in the winter greenhouse after being established in the fall. Ten accessions of *Erysimum* were harvested in the greenhouse in 2009. Seven of the accessions were established in fall 2008, two in 2006 and one in 2007. Only one of the ten accessions will need to be maintained into 2010 to obtain a sufficient amount of seed for standard distribution.

Linum:

Cultivated flax accessions are 99% available. One accession of *Linum usitatissimum* was planted and harvested in 2009. Wild flax accessions are 72% available. Five wild flax accessions were harvested in 2009; one from the field, three from populations maintained in the greenhouse, and one accession was harvested from populations in both the field and the greenhouse.

Cuphea:

Seeds are available for 94% of the accessions of seven species (*Cuphea calophylla*, *C. carthagenensis*, *C. lanceolata*, *C. lutea*, *C. toluicana*, *C. viscosissima*, *C. wrightii*) and the *Cuphea* hybrid accessions that have been part of the PSR23 breeding efforts by members of the National *Cuphea* Consortium for the agronomic development of *Cuphea* as a domestic source of mid-chain fatty acids. Over all, the *Cuphea* collection is 78% available. Six accessions were established in the field in 2009, four of which were harvested. Three accessions were transferred to the greenhouse for winter 2010 harvest.

Miscellaneous asters, *Vernonia*:

Overall, the miscellaneous asters are 24% available. In 2009, we harvested seeds from an accession of *Vernonia glabra* transferred to FGH-1 from the field in fall 2008.

Euphorbia:

In response to potential future interest in *Euphorbia lagascae* as a source of vernolic acid for the “green” building trade, in 2008 we began to increase the availability these accessions, the first field increases of *Euphorbia* at the NCRPIS since 1998. Seven accessions were successfully increased and made available. In 2009, ten accessions of *E. lagascae* were successfully increased and this species is now 56% available, an increase in availability of 7% over 2008. Overall, the *Euphorbia* collection is 36% available.

Distributions:

General statistics about oilseed collection distributions are presented in Table 3 in the appendix.

Helianthus:

Most requests in 2009 for sunflower accessions were for germplasm to support genetic or genomic-related research; however, requests for material to support disease-resistance and insect-tolerance evaluations commanded the largest number of accessions. Additional *Helianthus* inbred accessions were supplied to the research group continuing work on an NSF-funded association-mapping project begun in 2008. Accessions were also distributed in support of a number of breeding programs with interests in agronomic characteristics, including an ornamental sunflower breeding program.

Brassicaceae:

The largest proportion of orders of Brassicaceae accessions was used to supply material to support breeding programs, including programs to investigate environmental adaptability (winter hardiness, production in tropical highlands). Disease-resistance evaluations continue to be a major use of distributed material. In addition, portions of the Brassicaceae collection were distributed for phytoremediation research, bio-fumigant studies, and oil composition, biofuel and oil crop evaluation. The diversity in the Brassicaceae collection (262 taxa from 21 genera) supports a wide range of research purposes.

Linum:

The largest request for *Linum* accessions came from Plant Genetic Resources of Canada to allow them to complete increases of the World Flax Collection (NPGS collection) accessions and make them available to researchers. There were also two orders for material to support oil properties research as well as one order for research to develop perennial flax for inclusion in a new cropping system.

Cuphea:

Most *Cuphea* requests in 2009 were for material to support lipid metabolism and biofuel research. During 2008, several laboratories expressed interest in material not previously available for distribution from the NPGS for gene isolation and expression analysis of enzymes involved in the biosynthesis of specific mid-chain fatty acids. We began regeneration work for *C. painteri* and *C. averigera* var. *pulcherimia* and were able to distribute plant cuttings for both of these unusual species in 2009. Regeneration material is flowering and caged in the greenhouse; seeds should be available for distribution in 2010.

Euphorbia:

Euphorbia accessions were distributed during 2009 primarily to supply material for ornamental breeding work. Seven orders shipped in 2009 continues a trend of increase in *Euphorbia* requests, compared with an average of two orders per year from 2003 - 2007.

Miscellaneous asters:

The largest request for accessions from the NCRPIS miscellaneous asters group was to support ornamental breeding with the North American genus, *Vernonia*.

Research Activities:

General statistics about observations and images recorded for the collections are presented in Table 4 in the appendix.

Helianthus:

Woody Biomass Trait Analysis: We began field work in support of a multi-year, multi-location \$1.2 million research project evaluating woody traits in sunflower funded by the USDA/DOE (Ames is funded directly by the DOE). Overall grant management, a second field location, and all breeding and genetic/genomic work are taking place at the University of Georgia, Athens, GA. Chemical analyses are being handled by the National Renewable Energy Laboratory (NREL) in Golden, CO. The University of British Columbia, Vancouver is providing an additional field location. In addition to providing germplasm in support of the genetics and genomics portions of the project, in 2009, we grew 450 single-row field plots. Phenotypic observations (including leaf scans to determine area and stem volumetric measurements to determine specific density) were recorded (1624 plants) and samples harvested separately for DNA isolation (460 plants), chemical analyses (460 plants), biomass determinations (460 plants) and for an autonomous pollination sub-study (1552 plants). DNA extractions were completed in GA for leaf samples shipped from IA. First year biomass analyses have been completed and stem samples for wood analysis have been shipped to NREL. The long-term goal of this project is to identify genes associated with secondary xylem accumulation observed in accessions of *Helianthus argophyllus* (a wild annual species native to coastal southwest TX) and *H. niveus* ssp. *tephrodes* (a long-lived annual native to sand dunes in southeastern CA and northern Mexico) and move them into elite seed oil production lines resulting in material useful both for oilseed harvest and biomass production.

Disease resistance evaluations: Sclerotinia is the most important disease in sunflower production fields in northern North America. The pathology group at the USDA Sunflower Research Unit, Fargo, began field evaluations in 2008 to screen all untested cultivated sunflower accessions for response to this important disease, after Laura Marek identified all accessions lacking disease-evaluation data. An initial test group of 250 was selected, seed pre-treated in Ames with an experimental pesticide combination, and planted at three locations in 2008. Adverse weather conditions resulted in the loss of two of the three 2008 field locations, requiring re-testing in the field in 2009. We again treated the seeds with pesticide prior to shipment. The 20 best accessions from the surviving 2008 location were tested at additional sites in 2009. These field evaluations are labor intensive and are clearly subject to variable weather conditions. We are also participating in joint efforts with the Ames and Fargo pathology groups to develop a greenhouse screening method

that accurately predicts field response. This would help focus field-testing resources on only the most promising accessions. Initial test groups for developing the greenhouse screen were selected from wild collections. The 2009 testing effort focused on wild annual species, including *H. annuus*, *H. praecox* and *H. argophyllus*, one of the species being evaluated in the woody biomass research project described above. Greenhouse screening proceeded in Ames and more resistant material was advanced to field trials managed by Fargo. 2009 field results correlated well with the greenhouse data and, perhaps unexpectedly, for a number of accessions, plants were more resistant under field conditions. Tested *H. argophyllus* accessions averaged more than 80% resistant.

Brassicaceae:

Thlaspi agronomic characteristics: *Thlaspi arvense*, a Brassicaceae weed species commonly associated with agricultural production world-wide, has remarkable cold tolerance and interesting seed-oil characteristics. *Thlaspi arvense* completes its life cycle by late spring or early summer, suggesting use as a non-food or feed biodiesel component in a double-cropping system. In cooperation with the USDA-ARS New Crops Research Unit in Peoria, IL, during 2007 and 2008 we started to examine some of the agronomic characteristics of the *Thlaspi* germplasm in the NCRPIS collection and provide seed for further evaluations. In fall 2008, Ivan Ayala, an ISU Fulbright-sponsored graduate student from Colombia took over this project, which was expanded to include examination of *Camelina sativa* accessions in the NCRPIS collection, another member of the Brassicaceae with remarkable cold tolerance. Spring planting in 2009 was delayed due to wet field conditions, and *Thlaspi* accessions planted in May did not germinate well. However, *Camelina* accessions germinated quickly and well, and were harvested for oil analyses planned for spring 2010. *Thlaspi* evaluation plots were planted in early November, and the project will continue in 2010 and 2011.

Cuphea:

Fatty-acid biosynthetic enzyme gene isolation: We have been cooperating with Dr. Marna Yandea-Nelson, a scientist in Dr. Basil Nikolau's lab, ISU, to provide germplasm, field cage and greenhouse space for *Cuphea* plants being harvested for RNA and gene isolation. Drs. Yandea-Nelson and Nikolau are interested in enzymes involved in controlling preferential accumulation in seeds of specific mid-chain fatty acids, and various *Cuphea* species have unique seed fatty-acid profiles. We have been able to provide germplasm not available from any other source (notably *C. avigeria* var. *pulcherrima*, *C. painteri*, and *C. inflata*). The ISU group has begun gene isolation work and will continue field work with several additional *Cuphea* species in 2010.

Collection trips:

Laura Marek was the PI for one targeted collection trip, funded by the NPGS Plant Exchange Office (PEO), for wild *Helianthus* germplasm in 2009. In October, Laura met Dr. Gerald Seiler, Botanist, USDA Sunflower Research Group, Fargo in Des Moines, IA. They spent 13 days collecting wild sunflowers in the south central U.S., traveling more than 4600 miles across Kansas, Oklahoma, Arkansas and Missouri. The new collections represent the first time that germplasm for *H. salicifolius* will be available in quantities large enough to allow for standard distributions and the first time that material from the Arkansas subpopulation of *H. occidentalis* ssp.

plantagineus has been represented in the NPGS collection. In addition, new collections of *H. laetiflorus*, *H. pauciflorus* ssp. *pauciflorus*, and *H. silphioides* added significant regional diversity to the existing collection.

Professional Activities:

Training:

First aid and CPR training at the NCRPIS

Meetings and Presentations:

Helianthus: In January, Laura attended the 17th Annual Plant and Animal Genome Conference and participated in an afternoon meeting of the sunflower woody biomass trait analysis research group.

In June 2009, Laura attended the National Sunflower Association's Summer Seminar in Alexandria, MN and presented the *Helianthus* Germplasm Status Report to the Sunflower CGC prior to the start of the Summer Seminar.

NC7-RTAC: In August, Laura presented an update on the NCRPIS Oilseeds Project to the NC7-Regional Technical Advisory Committee at their annual meeting in Ames, IA.

Publications:

Griffiths, P.D., Marek, L.F., and Robertson, L.D. 2009. Identification of crucifer accessions from the NC-7 and NE-9 plant introduction collections that are resistant to black rot (*Xanthomonas campestris* pv. *campestris*) Races 1 and 4. HortScience 44(2):284-288.

Block, C.C., Gulya, T.J. and Marek, L.F. 2009. Evaluation of wild sunflower species for resistance to Sclerotinia stalk rot. Phytopathology 99:S13

Grant applications:

FY 2010 Southern US *Helianthus* collection trip proposal submitted to the NPGS PEO office, \$7163.

FY 2009 South Central US *Helianthus* collection trip proposal approved and funded \$7558.

FY2010 Plant Germplasm Evaluation proposal submitted, "Evaluation of *Thlaspi* and *Camelina* Accessions", \$15,000 (molecular analyses).

FY2009 Plant Germplasm Evaluation proposal approved and funded, "Evaluation of *Thlaspi* and *Camelina* Accessions," \$12,675 (agronomic analyses)

USDA/DOE Biomass Feedstock grant continuation funding approved: Genomics of Wood Formation and Cellulosic biomass Traits in Sunflower; Dr. Steve Knapp, UGA, principal investigator, \$1,200,00 total funding, of which ISU received \$150,000 as a separate DOE contract over three years (FY 2009 - 2011).

Genome Canada Genomics of Sunflower, \$11 million total funding, collaborator

Service Activities:

NCRPIS:

Laura serves on the NCRPIS Safety and Computer Committees.

Agronomy Departmental activities:

Laura continues to coordinate the monthly Agronomy Department Professional and Scientific staff meetings.

Plant Germplasm Operations Committee (PGOC):

Laura serves as a member of PGOC's *In situ* Conservation Subcommittee, GIS and Geo-referencing Subcommittee, and Molecular Subcommittee.

H. 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), *Daucus* (NC7-daucus), *Ocimum* (NC7-ocimum), and *Pastinaca* (NC7-parsnips). Statistics for accession numbers and availability for each site crop are found in the appendices in "Table 1: NCRPIS Accessions (Accs), Acquired, Available."

Acquisition:

In 2009, 104 new accessions were received and are listed by site crop in Table 1. The new accessions include two *Cucumis melo* (an old French cultivar from Michel Lachaume, Hip-Gnosis Seed Development, Canada; and 'Armenian Yard Long', from NCGRP), 10 *C. sativus* (cultivars used as disease differentials, from NCGRP), and 91 *Daucus* (collected in Tunisia by Drs. David Spooner and Philipp Simon, USDA-ARS, University of Wisconsin, Madison, WI).

Maintenance:

Data for vegetable crop regenerations attempted and number of accessions harvested in 2009 are summarized in the appendices in "Table 2: NCRPIS Accessions (accs) Germinated, Regenerated, Made Available, Backed Up."

Cucumis field regenerations included 65 new *C. melo* from Turkmenistan, nine *C. melo* with low seed quantities, one NSL-numbered *C. melo* requiring taxonomic verification and increase, one *C. sativus*, one *C. dipsaceus*, and one *C. ficifolius*. Of the 78 accessions planted for regeneration, four failed to germinate, 16 had no mature fruit before frost, 39 had insufficient fruit or seed production, and 19 accessions produced a sufficient quantity of seeds to be made available for distribution and backed up at NCGRP. The wet spring and early summer combined with the lack of heat units caused slow vine development and increased disease pressures resulting in poor plant performance and fruit development.

Cucurbita pepo field regenerations focused on accessions with low seed quantities or distribution lots 20+ years old. Twelve of 17 accessions were successfully regenerated. The five unsuccessful regenerations include two accessions that failed to germinate; one had a single plant which failed to set fruit, and two produced few fruit resulting in an insufficient seed increase. Twelve increases should be available for distribution after viability testing in April 2009.

Daucus regeneration efforts included both new accessions and old ones with low seed quantity or viability. Processing of the 2009 field cage and greenhouse harvests is still in progress. Preliminary assessment of the harvest indicated that two accessions failed to germinate, three accessions exhibited poor plant performance and seed production (one was determined to be male sterile), seven accessions produced low seed quantities, and 31 accessions have sufficient seed production to allow for distribution and NCGRP backup. In addition to regenerations in Ames, we received seed increases from R. Maxwell, Seminis Vegetable Seeds, Idaho (6 accessions), and R. Freeman, Nunhems, Oregon (3 accessions). Dr. Maxwell will be sent another 6 PI-numbered accessions for regeneration in 2010.

As NCRPIS accessions are regenerated, seed samples are sent to NCGRP for backup. Overall, 82% of the vegetable collections are backed up, which is above the station's average of 79%. Five of eight vegetable site-crops have 81% or more of their accessions backed up at NCGRP (Table 2). In addition to the backup samples sent to NCGRP, 293 accessions from the vegetable site-crops were sent to the Svalbard Global Seed Vault in Norway for backup in 2009.

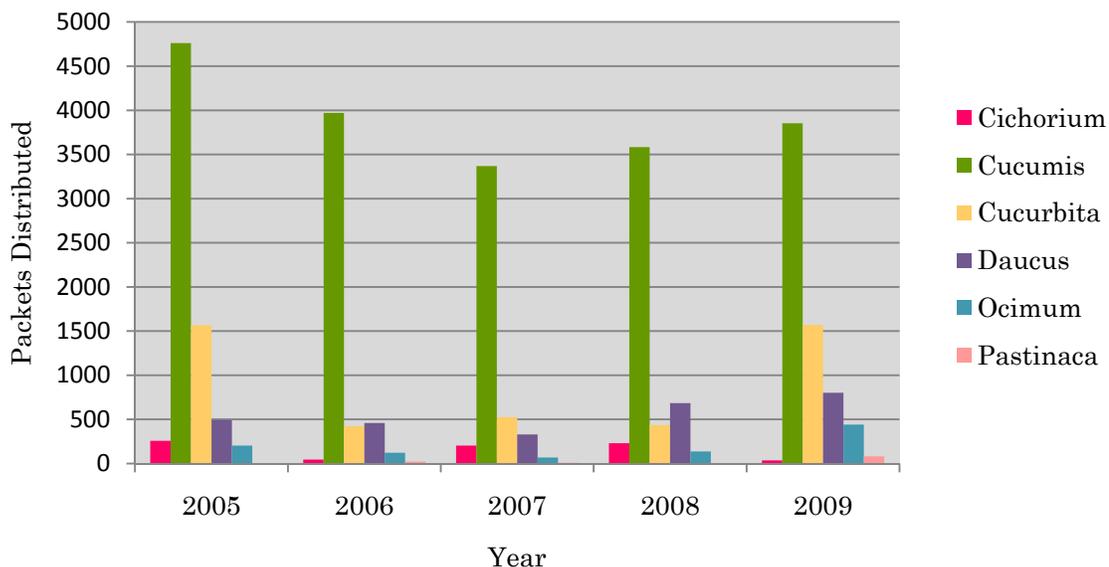
No vegetable accessions were inactivated in 2009.

In 2009, 720 germination tests (Table 2) were performed; 579 tests to monitor *Cucumis melo* distribution seed lot viabilities and 141 tests on seed increases from the 2008 regenerations.

Distribution:

Packet and accession distributions for the vegetable collections are summarized in the appendices in "Table 3A: External NCRPIS Distributions" and "Table 3B: Internal NCRPIS Distributions." In 2009, 6784 seed packets (items) involving 3915 accessions were distributed to fulfill 490 domestic and 56 foreign orders. A distribution history of the vegetable crops for the last five years can be found in the chart below and in the appendices in "Table 5: Five-Year Summary of NCRPIS Accession Orders by Crop."

5-yr Vegetable Distributions



All available *Cucumis sativus* accessions (1132 items) were distributed as one order for molecular analysis, and another order for all available *Cucurbita pepo* (799 items) was sent out for disease evaluation and breeding. Other large orders for *Cucumis*, *Cucurbita*, and *Daucus* were distributed for comparative genomics, oil analysis, and breeding.

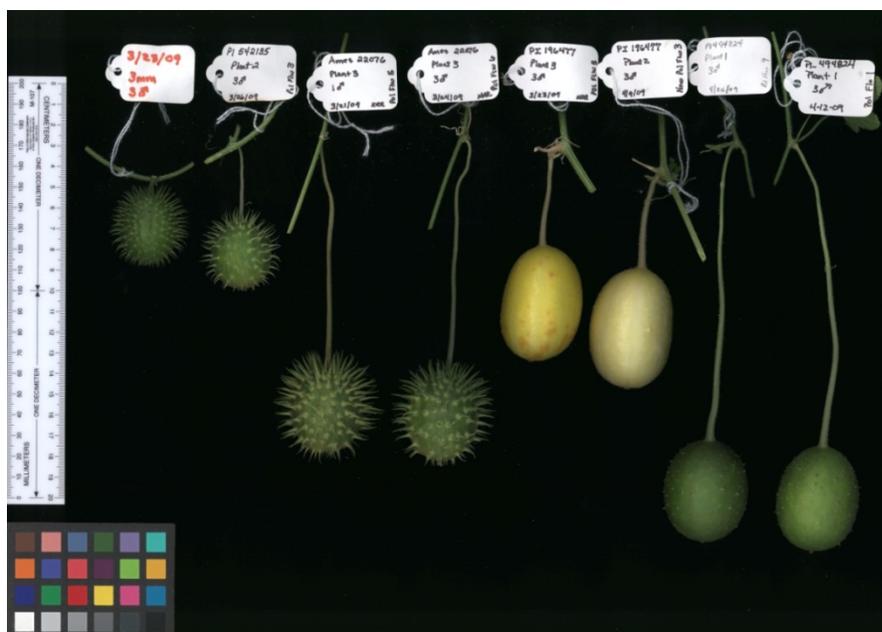
Characterization and Taxonomy:

Digital images, along with basic notes for taxonomic identification and accession characterization, were recorded during regeneration. Data for approximately 17 descriptors, primarily fruit descriptors, were recorded at harvest for *Cucumis* and *Cucurbita* accessions. Plant habit, flowering dates, and life-cycle notes were recorded for *Daucus*.

In May, 53 *Daucus* accessions were direct seeded into an observation field to verify taxonomy, collect characterization data, and capture digital images of plants, flowers, foliage, and roots for GRIN.

With the assistance of Dr. Mark Widrlechner (NCRPIS Horticulturist), taxonomic identities are reviewed and confirmed as each accession is regenerated or grown in observation plots. The 2009 re-identifications included nine *Cucumis* accessions to other species within *Cucumis*, and six *Daucus* to other *Daucus* species, subspecies, or varieties.

Dr. Widrlechner, Cindy Clark, and Kathy Reitsma (NCRPIS, Ames, IA), along with Dr. Joseph Kirkbride, Jr. (USDA-ARS, Beltsville, MD) submitted an article, "Length and Rapid Elongation of Pedicles of the Female Flowers of *Cucumis anguria* L.", for publication in the Cucurbit Genetics Cooperative Report. The article documents the variability of pedicle lengths encountered in *Cucumis anguria* germplasm, and revises the key published in Dr. Kirkbride's Biosystematic Monograph of the Genus *Cucumis* (Cucurbitaceae) describing this species. The report should be published in 2010.



Pedicel length variability in *Cucumis anguria*

Evaluation/Utilization:

Dr. Charles Block (Pathologist) continued to screen all *Cucurbita* and *Cucumis* seedlings grown for regeneration for the presence of Squash Mosaic Virus, by using ELISA protocols before seedlings are transplanted to the field. Seedling screening has been done since 1993. He also visually inspected all cucurbit field plantings for disease during the 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. Please refer to the Plant Pathology Project section of this report for more information.

The Pollinator Program and the Vegetable Program continued to collaborate on pollinator tests. In 2009, we updated our *Cucurbita* field-cage regeneration protocols to incorporate use of customized honey bee and bumblebee domicile-protection stands. These stands were evaluated in a 2008 study to determine whether they improved insect-pollinator performance within the cages. Also, preliminary results of an informal 2009 study to improve alfalfa leaf cutter (ALC) bee activity through the use of multiple ALC domiciles in *Cucumis* cages suggests further investigation is warranted when resources are available. For more information on this work, please refer to the Entomology section of the annual report.

Publications/Posters:

Widrechner, Mark P., Kathleen R. Reitsma, Lucinda D. Clark, and Joseph H. Kirkbride, Jr. 2009. Length and rapid elongation of pedicels of the female flowers of *Cucumis anguria* L. Cucurbit Genetics Cooperative Report 31/32:36-40 and back cover.

Plans for 2010:

Regenerations:

Thirty-one accessions of *Daucus* were started in the greenhouse in October 2009, and we may start an additional 15 annual *Daucus* in the greenhouse in April 2010 for the 2010 field cages. We will attempt to regenerate approximately 100 *Cucumis* accessions, focusing on accessions that produced insufficient seed quantities in 2009. We will also regenerate approximately 15 *Cucurbita* accessions having low seed quantities. Regenerations of wild *Cucumis* species and hard-to-handle *Cucumis* will continue in the greenhouse as time, space, and other resources permit.

Germinations:

Viability tests will be performed on the 2009 cucurbit regeneration seed lots in April 2010 and on the 2009 *Daucus* regeneration seed lots in the summer of 2010. Ten-year re-germination testing will be done as resources allow.

Characterization:

A 2-year observation planting of selected *Daucus* accessions will be direct seeded to the field in the spring of 2010 for characterization and taxonomic verification. This plot will be in collaboration with Drs. David Spooner and Philipp Simon (USDA-ARS, University of Wisconsin, Madison, WI) to evaluate *D. carota* diversity, and to evaluate some of the newly collected *Daucus* populations from the 2009 Tunisia collection trip. The resulting data and images will be loaded into GRIN. These data and images may prove useful in Dr. Spooner's work to develop a monograph for the genus *Daucus*.



Daucus root image



Daucus root coin image



Daucus leaf scan

Review of accession passport data will continue on the cucurbit collections in preparation for assigning PI numbers to many of the Ames-numbered accessions in the collections (414 *Cucumis*, 91 *Cucurbita*). Labeling embedded in digital images of these accessions will be updated with the new PI numbers before they are loaded to GRIN.

Evaluation:

Collaboration continues on improving the year-round cage and insect-pollinator program for regenerating vegetable crops.

The Plant Pathology Project will continue to collaborate in monitoring 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 of cucurbits. They will also continue the greenhouse survey of the *Cucumis melo* distribution lots for the presence of *Acidovorax avenae* ssp. *citrulli*.

I. Research Leader Activities (C. Gardner)

Administration and Leadership Activities:

C. Gardner administers the five-year project-plan objectives for the USDA-ARS Plant Introduction Research Unit's two CRIS Projects, Plant Introduction Research and the Germplasm Enhancement of Maize (GEM) Project, and contributes to the coordination and execution of activities which support those objectives. Gardner serves as the Coordinator of the Hatch-funded Multistate NC7 Project, and as a member of the National Plant Germplasm Coordinating Committee, dedicated to increasing awareness and enhancing the operations of the NPGS across its components.

About 30% of her time in 2009 was devoted to assisting GRIN-Global System development team members. Funded in part by the Global Crop Diversity Trust via a grant from the Gates Foundation, and in part by the USDA-ARS through dedication of key personnel, the project's objective is to develop a rational genebank information management system which will be free of recurring license fees, database neutral, and can be used on either stand-alone or networked computers by any genebank in the world. Source code developed as a result of the project will be

made freely available. The GRIN-Global system will be implemented internationally in late 2010, and content of the NPGS' legacy GRIN system will be migrated to the GRIN-Global System in the U.S. in the future.

A Release Candidate was released to selected genebank personnel for beta testing at the end of 2009. During the first half of 2010, a new public interface prototype will be available for testing. Bioversity International provides the expertise for translation of the interface into several languages, development of training and documentation materials, and training of international genebank personnel.

Pete Cyr, our Software Applications and Network Systems Information Specialist, serves as the project leader. Together with Mark Millard, our maize curator who serves as systems analyst for the project; Lisa Burke, our seed storage manager who serves as a primary beta tester; Brock Weaver, a contract software developer employed by Bioversity (Ames); Rachelle Little, a contract web developer employed by Bioversity (Ames); Joe Postman, pathologist with the ARS Corvallis, OR, National Clonal Germplasm Repository as a tester; the ARS GRIN Database Management Unit (DBMU) personnel; National Program Leader (and Project PI) Peter Bretting; and our Global Crop Diversity Trust and Bioversity partners, our development efforts will be primarily devoted to this critically important project for two more years.

Research Activities:

Graduate student Ivan Ayala-Diaz, a Fulbright Fellow from Colombia, is conducting his Ph.D. research on *Thlaspi* and *Camelina* under the guidance of Dr. Mark Westgate, ISU, and Dr. Gardner, and in collaboration with NCRPIS Oilseeds Curator, Dr. Laura Marek.

Another graduate student, Adam Vanous, is working on an M.S. project dealing with GEM Project germplasm and generation of dihaploid lines from introgressed maize racial materials.

Professional Interactions:

CSSA C8 Division: Chair-elect for 2011.

Year 2009 Table 1.

NCRPIS Accessions (Accs), Acquired, Available

01/01/2009 to 12/31/2009

CURATOR	GENUS_CROP	Number			Percent		Percent Avail Last Year*
		Number Accs	Accs Acquired	Percent Acquired	Number Available	Percent Available	
Brenner	NC7-amaranth	3343	1	0	3191	95	96
	NC7-celosia	55	0	0	27	49	49
	NC7-echinochloa	306	0	0	248	81	81
	NC7-grasses	126	3	2	76	60	62
	NC7-legumes	234	1	0	108	46	46
	NC7-melilotus	997	14	1	768	77	77
	NC7-panicum	945	0	0	912	97	96
	NC7-perilla	24	0	0	23	96	88
	NC7-quinoa	351	9	3	227	65	60
	NC7-setaria	1014	0	0	912	90	90
	NC7-spinach	402	1	0	347	86	86
	NC7-umbels	1135	37	3	627	55	55
	Total:	8932	66	1	7466	84	84
Marek	NC7-asters	362	10	3	87	24	22
	NC7-brassica	2002	8	0	1810	90	90
	NC7-crucifers	1142	5	0	970	85	83
	NC7-crucifers.pvp	1	0	0	0	0	0
	NC7-cuphea	639	0	0	499	78	79
	NC7-euphorbia	208	0	0	75	36	33
	NC7-flax	2834	0	0	2818	99	99
	NC7-flax.wilds	119	5	4	86	72	70
	NC7-sun.cults	1736	18	1	1655	95	94
	NC7-sun.wilds.ann	1377	12	1	1293	94	93
	NC7-sun.wilds.per	831	60	7	501	60	59
	NC7-sun.wilds.sp	11	0	0	5	45	45
		Total:	11262	118	1	9799	87
Millard	NC7-corn.kin	34	0	0	6	18	18
	NC7-maize.gems	107	24	22	103	96	100
	NC7-maize.inb	2397	9	0	1770	74	76
	NC7-maize.pop	17078	4	0	11086	65	64
	NC7-maize.pvp	208	39	19	191	92	91
	NC7-maize.wilds	376	29	8	89	24	18
	Zea.totals	20166	105	1	13239	66	65
	Total:	20200	105	1	13245	66	65
Qu	NC7-medicinals	489	40	8	310	63	64
	Total:	489	40	8	310	63	64
Reitsma	NC7-chicory	276	0	0	229	83	83
	NC7-cucumis.cucs	1374	10	1	1291	94	94
	NC7-cucumis.melo	3194	2	0	2274	71	72
	NC7-cucumis.wilds	322	0	0	160	50	44
	NC7-cucurbita	993	0	0	770	78	81
	NC7-daucus	1218	92	8	977	80	84
	NC7-ocimum	98	0	0	91	93	93
	NC7-parsnips	70	0	0	51	73	73
		Total:	7545	104	1	5843	77
Widrechner	NC7-mints	153	9	6	107	70	72
	NC7-ornamentals	2337	236	10	1070	46	45
	Total:	2490	245	10	1177	47	47
NCRPIS Total:		50918	678	1	37840	74	74

Year 2009 Table 2.

NCRPIS Accessions (Accs) Germinated, Regenerated, Made Available, Backed Up

01/01/2009 to 12/31/2009

CURATOR	GENUS_CROP	Number Accs	Number Accs Germed	Percent Accs Germed	Number Attempted Regen	Number Harvested Regen	Number Perennial Perm	Number Perennial (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	3343	461	8	11	15	0	0	3	0	304	3212	96	
	NC7-eclosia	55	4	7	9	6	0	0	0	0	0	30	55	
	NC7-echinochloa	306	29	5	14	18	0	0	0	0	163	262	86	
	NC7-grasses	126	2	1	2	2	0	0	2	0	0	81	64	
	NC7-legumes	234	3	1	2	1	0	0	1	0	0	175	75	
	NC7-melilotus	997	12	1	31	0	0	0	12	0	12	843	85	
	NC7-panicum	945	2	0	11	8	0	0	8	0	0	915	97	
	NC7-perilla	24	3	13	1	1	0	0	3	0	3	23	96	
	NC7-quinoo	351	44	11	66	59	0	0	30	0	38	249	71	
	NC7-setaria	1014	43	4	7	6	0	0	4	0	636	956	94	
	NC7-spinach	402	1	0	34	3	0	0	1	0	0	374	93	
	NC7-umbels	1135	26	1	57	39	0	0	21	0	11	664	59	
	Total:		8932	630	7	245	158	0	0	85	0	1167	7784	87
	Marek	NC7-asters	362	5	1	1	0	0	0	8	0	1	98	27
		NC7-brassica	2002	43	2	34	32	0	0	40	8	26	1957	98
NC7-crucifers		1142	65	6	12	26	1	0	40	31	26	996	87	
NC7-crucifers.pvp		1	0	0	0	0	0	0	0	0	0	1	100	
NC7-cuphea		639	10	2	6	4	0	0	10	0	5	583	91	
NC7-euphorbia		208	6	3	10	10	0	0	7	0	5	78	38	
NC7-flax		2834	15	1	1	0	0	0	8	0	6	2832	100	
NC7-flax.wilds		119	9	8	7	5	0	0	8	3	8	86	72	
NC7-sun.cults		1736	39	2	45	40	0	0	61	0	36	1671	96	
NC7-sun.wilds.ann		1377	30	2	52	28	0	0	29	0	25	1306	95	
NC7-sun.wilds.per		831	33	4	78	64	0	0	58	0	51	507	61	
NC7-sun.wilds.sp		11	0	0	6	1	0	0	0	0	0	5	45	
Total:			11262	255	2	252	210	1	0	269	42	189	10120	90
Millard		NC7-corn.kin	34	0	0	1	3	1	0	0	0	0	8	24
		NC7-maize.gems	107	21	20	4	4	0	0	21	0	21	46	43
	NC7-maize.inb	2397	142	6	218	214	0	0	119	0	25	1531	64	
	NC7-maize.pop	17078	1040	6	72	68	0	0	155	0	13	13104	77	
	NC7-maize.pvp	208	5	2	70	70	0	0	53	0	12	208	100	
	NC7-maize.wilds	376	1	0	0	1	0	1	29	0	0	44	12	
	Zea.totals	20166	1209	6	364	357	0	1	377	0	71	14933	74	
	Total:	20200	1209	6	365	360	1	1	377	0	71	14941	74	
	Qu	NC7-medicinals	489	54	11	61	32	41	30	21	0	14	318	65
		NC7-chicory	276	1	0	0	0	0	0	0	0	0	243	88
		NC7-eucumis.cucs	1374	28	2	2	2	0	0	32	0	239	1298	94
		NC7-eucumis.melo	3194	590	12	76	59	0	0	10	0	7	2526	79
		NC7-eucumis.wilds	322	19	6	6	18	0	0	20	0	14	159	49
		NC7-eucurbita	993	15	2	17	14	0	0	16	0	13	811	82
		NC7-daucus	1218	66	5	46	46	0	0	83	0	46	984	81
NC7-ocimum		98	0	0	0	0	0	0	1	0	25	91	93	
NC7-parsnips		70	0	0	0	0	0	0	0	0	0	47	67	
Total:		7545	719	10	147	139	0	0	162	0	344	6159	82	
Widriechner		NC7-mints	153	5	3	0	8	0	0	6	0	4	113	74
		NC7-ornamentals	2337	50	2	26	110	80	9	123	0	59	939	40
		Total:	2490	55	2	26	118	80	9	129	0	63	1052	42
NCRPIS Total:		50918	2922	6	1096	1017	123	40	1043	42	1848	40374	79	

Year 2009 Table 3A. External NCRPIS Distributions

01/01/2009 to 12/31/2009

CURATOR	GENUS_CROP	External Domestic Distributions					Foreign Distributions					External Domestic and Foreign Distributions				
		Number Accs in Collection	Number Accs	Number Orders	Number Recipients	Number Items	Number Accs	Number Orders	Number Recipients	Number Items	Number Accs	Number Orders	Number Recipients	Number Items		
Brenner	NC7-amaranth	3343	287	60	56	411	341	11	11	463	540	71	67	874		
	NC7-celosia	55	16	11	10	22	3	2	2	3	16	13	12	25		
	NC7-echinochloa	306	38	7	7	40	20	2	2	20	51	9	9	60		
	NC7-grasses	126	1	1	1	1	0	0	0	0	1	1	1	1		
	NC7-legumes	234	21	12	11	28	3	3	3	4	22	15	14	32		
	NC7-melilotus	997	147	20	16	176	97	3	3	100	218	23	19	276		
	NC7-panicum	945	139	14	14	197	30	6	6	31	159	20	20	228		
	NC7-perilla	24	6	11	11	18	23	3	3	46	23	14	14	64		
	NC7-quinoo	351	147	43	43	249	160	14	11	326	223	57	54	575		
	NC7-setaria	1014	59	13	13	88	7	2	2	7	65	15	15	95		
	NC7-spinach	402	105	29	26	154	345	0	0	373	351	29	26	527		
	NC7-umbels	1135	191	57	53	259	199	7	7	201	276	64	60	460		
	Total:	8932	1157	278	261	1643	1228	53	50	1574	1945	331	311	3217		
	Marek	NC7-asters	362	31	13	10	40	1	1	1	1	31	14	11	41	
		NC7-brassica	2002	938	63	58	1283	351	19	18	413	1037	82	76	1696	
NC7-crucifers		1142	207	44	43	247	257	12	11	308	389	56	54	555		
NC7-crucifers.pvp		1	0	0	0	0	0	0	0	0	0	0	0	0		
NC7-cuphea		639	104	23	18	127	10	4	4	11	107	27	22	138		
NC7-euphorbia		208	29	8	6	35	0	0	0	0	29	8	6	35		
NC7-flax		2834	32	20	17	40	80	2	2	80	111	22	19	120		
NC7-flax.wilds		119	23	8	7	27	9	1	1	9	30	9	8	36		
NC7-sun.cults		1736	465	81	70	566	152	16	13	192	550	97	83	758		
NC7-sun.wilds.ann		1377	122	29	25	162	261	17	16	314	349	46	41	476		
NC7-sun.wilds.per		831	177	23	22	220	110	7	7	126	223	30	29	346		
NC7-sun.wilds.sp		11	0	0	0	0	0	0	0	0	0	0	0	0		
Total:		11262	2128	312	276	2747	1231	79	73	1454	2856	391	349	4201		
Millard		NC7-corn.kin	34	7	8	8	16	5	2	2	5	7	10	10	21	
		NC7-maize.gems	107	84	22	15	283	39	3	3	41	87	25	18	324	
	NC7-maize.inb	2397	852	264	195	2708	394	46	42	624	916	310	237	3332		
	NC7-maize.pop	17078	2766	298	259	3796	121	23	19	134	2810	321	278	3930		
	NC7-maize.pvp	208	194	221	111	3219	151	14	13	510	194	235	124	3729		
	NC7-maize.wilds	376	149	57	49	274	22	7	6	39	149	64	55	313		
	Zea.totals	20166	4045	692	457	10280	727	76	65	1348	4156	768	522	11628		
	Total:	20200	4052	700	465	10296	732	78	67	1353	4163	778	532	11649		
	Qu	NC7-medicinals	489	136	60	48	217	48	4	4	50	154	64	52	267	
		Total:	489	136	60	48	217	48	4	4	50	154	64	52	267	
		Reitsma	NC7-chicory	276	26	19	18	35	0	0	0	26	19	18	35	
			NC7-cucumis.cucs	1374	1201	101	96	1826	186	15	13	220	1217	116	109	2046
			NC7-cucumis.melo	3194	1028	128	118	1362	179	14	13	211	1099	142	131	1573
			NC7-cucumis.wilds	322	106	12	12	163	63	10	9	72	128	22	21	235
			NC7-cucurbita	993	807	106	100	1316	209	8	8	255	817	114	108	1571
NC7-daucus			1218	475	78	74	753	48	5	5	49	491	83	79	802	
NC7-ocimum			98	91	35	33	322	91	4	4	120	91	39	37	442	
NC7-parsnips			70	46	11	11	80	0	0	0	0	46	11	11	80	
Total:			7545	3780	490	462	5857	776	56	52	927	3915	546	514	6784	
Widrechner			NC7-mints	153	91	31	29	160	19	2	2	19	92	33	31	179
			NC7-ornamentals	2337	366	98	84	544	62	12	11	63	390	110	95	607
			Total:	2490	457	129	113	704	81	14	13	82	482	143	126	786
NCRPIS Total:			50918	11710	1260	889	21464	4096	227	192	5440	13515	1487	1081	26904	

Year 2009 Table 3B. Internal NCRPIS Distributions

01/01/2009 to 12/31/2009

NC7 Related (# Accs)

Seed Storage Maintenance

CURATOR	GENUS_CROP	Number Accs	Backed Up	Germated	Obs	Regen	Path Test	Total	# Distinct Accs for NC7 Orders	# Accs Stored	# Accs Ct Rev	
Brenner	NC7-amaranth	3343	304	461	88	52	0	905	596	2	243	
	NC7-celosia	55	0	4	0	9	0	13	12	2	1	
	NC7-echinochloa	306	163	29	12	22	0	226	203	2	37	
	NC7-grasses	126	0	2	2	3	0	7	6	3	0	
	NC7-legumes	234	0	3	1	2	0	6	5	1	0	
	NC7-melilotus	997	12	12	37	31	0	92	80	26	0	
	NC7-panicum	945	0	2	22	11	0	35	35	0	0	
	NC7-perilla	24	3	3	0	2	0	8	3	3	3	
	NC7-quinoa	351	38	44	5	66	0	153	72	74	17	
	NC7-sectaria	1014	636	43	40	18	50	787	721	4	139	
	NC7-spinach	402	0	1	4	34	0	39	38	1	31	
	NC7-umbels	1135	11	26	8	48	0	93	80	40	10	
	Total:		8932	1167	630	219	298	50	2364	1851	158	481
	Marek	NC7-asters	362	1	5	0	0	0	6	6	11	0
		NC7-brassica	2002	26	48	1428	34	0	1536	1461	53	72
NC7-crucifers		1142	26	66	62	19	0	173	144	53	57	
NC7-crucifers.pvp		1	0	0	0	0	0	0	0	0	0	
NC7-cuphea		639	5	10	0	6	0	21	16	11	9	
NC7-euphorbia		208	5	6	0	10	0	21	17	7	2	
NC7-flax		2834	6	215	2	1	0	224	218	8	264	
NC7-flax.wilds		119	8	9	0	8	0	25	17	17	4	
NC7-sun.cults		1736	36	38	11	42	0	127	88	58	169	
NC7-sun.wilds.ann		1377	25	31	96	39	0	191	158	40	13	
NC7-sun.wilds.per		831	51	33	2	58	0	144	122	121	7	
NC7-sun.wilds.sp		11	0	0	4	6	0	10	10	0	0	
Total:			11262	189	461	1605	223	0	2478	2257	379	597
Millard		NC7-corn.kin	34	0	0	0	0	0	0	0	0	0
		NC7-maize.gems	107	21	20	17	0	20	78	38	21	7
	NC7-maize.inb	2397	25	117	348	21	152	663	588	249	190	
	NC7-maize.pop	17078	13	476	808	14	37	1348	1307	222	967	
	NC7-maize.pvp	208	12	2	133	12	2	161	145	53	60	
	NC7-maize.wilds	376	0	1	0	2	2	5	4	30	10	
	Zea.totals	20166	71	616	1306	49	213	2255	2082	575	1234	
	Total:	20200	71	616	1306	49	213	2255	2082	575	1234	
	Total:	489	14	62	8	61	0	145	117	40	69	
	Reitsma	NC7-chicory	276	0	1	1	0	0	2	2	0	0
NC7-cucumis.cues		1374	239	28	0	2	0	269	241	31	108	
NC7-cucumis.melo		3194	7	590	1	76	276	950	891	13	435	
NC7-cucumis.wilds		322	14	19	6	7	0	46	30	21	12	
NC7-cucurbita		993	13	16	2	17	0	48	33	17	43	
NC7-daucus		1218	46	66	58	40	0	210	197	109	99	
NC7-ocimum		98	25	0	4	0	0	29	28	1	6	
NC7-parsnips		70	0	0	1	0	0	1	1	0	43	
Total:		7545	344	720	73	142	276	1555	1423	192	746	
Widrechner		NC7-mints	153	4	5	0	0	0	9	5	8	1
	NC7-ornamentals	2337	59	39	11	56	0	165	139	197	66	
	Total:	2490	63	44	11	56	0	174	144	205	67	
NCRPIS Total:	50918	1848	2533	3222	829	539	8971	7874	1549	3194		

Year 2009 Table 4. NCRPIS Accessions (Accs) Observations (Obs) in GRIN, Images in GRIN

01/01/2009 to 12/31/2009

CURATOR	GENUS_CROP	Number Accs	Number Accs Obs Trials	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 years)	Number Accs Imaged	Number Acc Images in GRIN for Year	Number Acc Images in GRIN (all years)	
Brenner	NC7-amaranth	3343	88	50	21	33	3324	100	0	397	
	NC7-celosia	55	0	0	0	3	9	5	0	8	
	NC7-echinocloa	306	12	0	0	0	294	15	0	23	
	NC7-grasses	126	2	0	0	0	11	2	0	12	
	NC7-legumes	234	1	0	0	0	88	2	0	5	
	NC7-melilotus	997	37	1456	349	973	975	1	0	37	
	NC7-panicum	945	22	0	0	9	938	25	0	27	
	NC7-perilla	24	0	0	0	0	0	1	0	1	
	NC7-quinoa	351	5	0	0	14	245	50	0	41	
	NC7-setaria	1014	40	0	0	6	995	30	0	30	
	NC7-spinach	402	4	0	0	1	401	3	0	1	
	NC7-umbels	1135	8	1	1	65	227	25	1	13	
	Total:	8932	219	1507	371	1104	7507	259	1	595	
	Marek	NC7-asters	362	0	0	0	0	4	1	0	9
		NC7-brassica	2002	1428	46	41	69	1901	9	0	332
		NC7-crucifers	1142	62	50	43	57	821	0	0	334
		NC7-crucifers.pvp	1	0	0	0	0	1	0	0	0
NC7-euphea		639	0	2	2	10	367	3	2	10	
NC7-euphorbia		208	0	0	0	0	0	0	0	0	
NC7-flax		2834	2	46	8	6	2825	0	1	1	
NC7-flax.wilds		119	0	0	0	21	82	0	0	6	
NC7-sun.cults		1736	11	1933	115	35	1657	54	36	60	
NC7-sun.wilds.ann		1377	96	539	35	52	1264	42	28	49	
NC7-sun.wilds.per		831	2	898	63	149	550	122	54	104	
NC7-sun.wilds.sp		11	4	0	0	0	6	0	0	0	
Total:		11262	1605	3514	307	399	9478	231	121	905	
Millard	NC7-corn.kin	34	0	0	0	0	0	0	0	0	
	NC7-maize.gems	107	17	121	31	9	103	20	31	103	
	NC7-maize.inb	2397	348	1798	158	870	2147	226	26	588	
	NC7-maize.pop	17078	808	7235	522	1029	14452	247	36	4180	
	NC7-maize.pvp	208	133	1059	81	118	204	76	72	169	
	NC7-maize.wilds	376	0	29	29	0	264	30	0	107	
	Zea.totals	20166	1306	10242	821	2026	17170	599	165	5147	
	Total:	20200	1306	10242	821	2026	17170	599	165	5147	
	Qu	NC7-medicinals	489	8	4	4	262	291	18	3	269
		Total:	489	8	4	4	262	291	18	3	269
Reitsma		NC7-chicory	276	1	27	27	204	276	0	27	231
		NC7-cucumis.cues	1374	0	14	14	165	1363	2	14	872
		NC7-cucumis.melo	3194	1	11	11	160	3112	59	11	458
		NC7-cucumis.wilds	322	6	0	0	10	287	22	0	30
		NC7-cucurbita	993	1	150	97	987	987	16	97	97
		NC7-daucus	1218	58	0	0	304	1124	104	0	1
		NC7-ocimum	98	4	0	0	19	98	0	0	0
		NC7-parsnips	70	1	0	0	70	70	0	0	0
	Total:	7545	72	202	149	1919	7317	203	149	1689	
	Widrechner	NC7-mints	153	0	0	0	7	26	5	0	31
NC7-ornamentals		2337	16	220	150	254	590	139	150	622	
Total:	2490	16	220	150	261	616	144	150	653		
NCRPIS Total:	50918	3226	15689	1802	5971	42379	1454	589	9258		

CURATOR	GENUS_CROP	TIME_PERIOD	Number Orders	Number Recipients	Number Items Distributed	Number Accessions Distributed
Brenner	NC7-amaranth	01/01/2005 - 12/31/2005	59	53	539	228
		01/01/2006 - 12/31/2006	59	53	3230	2630
		01/01/2007 - 12/31/2007	56	50	532	310
		01/01/2008 - 12/31/2008	52	51	446	291
		01/01/2009 - 12/31/2009	71	67	874	540
		Total:	297	274	5621	3999
	NC7-celosia	01/01/2005 - 12/31/2005	5	5	7	6
		01/01/2006 - 12/31/2006	9	8	31	21
		01/01/2007 - 12/31/2007	8	8	35	21
		01/01/2008 - 12/31/2008	4	4	9	8
01/01/2009 - 12/31/2009		13	12	25	16	
	Total:	39	37	107	72	
NC7-echinochloa	01/01/2005 - 12/31/2005	5	5	58	56	
	01/01/2006 - 12/31/2006	19	16	49	24	
	01/01/2007 - 12/31/2007	5	5	15	14	
	01/01/2008 - 12/31/2008	5	4	13	12	
	01/01/2009 - 12/31/2009	9	9	60	51	
	Total:	43	39	195	157	
NC7-grasses	01/01/2005 - 12/31/2005	1	1	3	3	
	01/01/2006 - 12/31/2006	7	6	9	7	
	01/01/2007 - 12/31/2007	2	2	6	5	
	01/01/2008 - 12/31/2008	2	2	5	5	
	01/01/2009 - 12/31/2009	1	1	1	1	
	Total:	13	12	24	21	
NC7-legumes	01/01/2005 - 12/31/2005	7	6	28	24	
	01/01/2006 - 12/31/2006	6	6	15	11	
	01/01/2007 - 12/31/2007	2	2	9	9	
	01/01/2008 - 12/31/2008	11	10	86	78	
	01/01/2009 - 12/31/2009	15	14	32	22	
	Total:	41	38	170	144	
NC7-melilotus	01/01/2005 - 12/31/2005	16	14	83	73	
	01/01/2006 - 12/31/2006	12	10	56	49	
	01/01/2007 - 12/31/2007	12	11	41	27	
	01/01/2008 - 12/31/2008	20	15	411	268	
	01/01/2009 - 12/31/2009	23	19	276	218	
	Total:	83	69	867	635	
NC7-panicum	01/01/2005 - 12/31/2005	11	11	80	68	
	01/01/2006 - 12/31/2006	22	21	77	42	
	01/01/2007 - 12/31/2007	11	10	21	20	
	01/01/2008 - 12/31/2008	18	18	150	115	
	01/01/2009 - 12/31/2009	20	20	228	159	
	Total:	82	80	556	404	
NC7-perilla	01/01/2005 - 12/31/2005	11	11	74	22	
	01/01/2006 - 12/31/2006	14	14	102	22	
	01/01/2007 - 12/31/2007	6	6	21	14	
	01/01/2008 - 12/31/2008	5	5	44	19	
	01/01/2009 - 12/31/2009	14	14	64	23	
	Total:	50	50	305	100	
NC7-quinoa	01/01/2005 - 12/31/2005	32	30	302	138	
	01/01/2006 - 12/31/2006	37	31	401	196	
	01/01/2007 - 12/31/2007	40	37	307	128	
	01/01/2008 - 12/31/2008	50	45	296	132	
	01/01/2009 - 12/31/2009	57	54	575	223	
	Total:	216	197	1881	817	

	NC7-setaria	01/01/2005 - 12/31/2005	10	9	150	125
		01/01/2006 - 12/31/2006	23	21	227	177
		01/01/2007 - 12/31/2007	16	16	317	225
		01/01/2008 - 12/31/2008	24	24	316	249
		01/01/2009 - 12/31/2009	15	15	95	65
	Total:		88	85	1105	841
	NC7-spinach	01/01/2005 - 12/31/2005	12	12	399	370
		01/01/2006 - 12/31/2006	26	26	1619	377
		01/01/2007 - 12/31/2007	19	18	1196	374
		01/01/2008 - 12/31/2008	26	23	668	361
		01/01/2009 - 12/31/2009	34	31	527	351
	Total:		117	110	4409	1833
	NC7-umbels	01/01/2005 - 12/31/2005	27	25	239	215
		01/01/2006 - 12/31/2006	36	33	189	136
		01/01/2007 - 12/31/2007	30	29	199	164
01/01/2008 - 12/31/2008		30	29	463	313	
01/01/2009 - 12/31/2009		64	60	460	276	
Total:		187	176	1550	1104	
Brenner Total:			1256	1167	16790	10127
Marek	NC7-asters	01/01/2005 - 12/31/2005	9	9	73	70
		01/01/2006 - 12/31/2006	14	12	61	54
		01/01/2007 - 12/31/2007	7	6	16	16
		01/01/2008 - 12/31/2008	14	13	30	20
		01/01/2009 - 12/31/2009	14	11	41	31
	Total:		58	51	221	191
	NC7-brassica	01/01/2005 - 12/31/2005	56	54	1688	1286
		01/01/2006 - 12/31/2006	70	56	2532	1421
		01/01/2007 - 12/31/2007	64	49	1754	1047
		01/01/2008 - 12/31/2008	74	64	1866	1233
		01/01/2009 - 12/31/2009	82	76	1696	1037
	Total:		346	299	9536	6024
	NC7-crucifers	01/01/2005 - 12/31/2005	52	48	1486	497
		01/01/2006 - 12/31/2006	34	32	358	208
		01/01/2007 - 12/31/2007	48	43	839	412
01/01/2008 - 12/31/2008		59	50	658	520	
01/01/2009 - 12/31/2009		56	54	555	389	
Total:		249	227	3896	2026	
NC7-crucifers.pvp	01/01/2005 - 12/31/2005	0	0	0	0	
	01/01/2006 - 12/31/2006	0	0	0	0	
	01/01/2007 - 12/31/2007	0	0	0	0	
	01/01/2008 - 12/31/2008	0	0	0	0	
	01/01/2009 - 12/31/2009	0	0	0	0	
Total:		0	0	0	0	
NC7-cuphea	01/01/2005 - 12/31/2005	20	13	451	277	
	01/01/2006 - 12/31/2006	21	16	337	254	
	01/01/2007 - 12/31/2007	19	10	720	507	
	01/01/2008 - 12/31/2008	18	15	98	81	
	01/01/2009 - 12/31/2009	27	22	138	107	
Total:		105	76	1744	1226	
NC7-euphorbia	01/01/2005 - 12/31/2005	1	1	1	1	
	01/01/2006 - 12/31/2006	4	4	15	13	
	01/01/2007 - 12/31/2007	4	3	10	8	
	01/01/2008 - 12/31/2008	7	6	85	83	
	01/01/2009 - 12/31/2009	8	6	35	29	
Total:		24	20	146	134	

NC7-flax	01/01/2005 - 12/31/2005	14	14	1677	1441
	01/01/2006 - 12/31/2006	19	18	1284	1199
	01/01/2007 - 12/31/2007	8	8	60	56
	01/01/2008 - 12/31/2008	16	16	243	230
	01/01/2009 - 12/31/2009	22	19	120	111
Total:		79	75	3384	3037
NC7-flax.wilds	01/01/2005 - 12/31/2005	6	6	34	30
	01/01/2006 - 12/31/2006	8	8	138	65
	01/01/2007 - 12/31/2007	4	3	19	19
	01/01/2008 - 12/31/2008	6	6	24	22
	01/01/2009 - 12/31/2009	9	8	36	30
Total:		33	31	251	166
NC7-sun.cults	01/01/2005 - 12/31/2005	62	46	1635	789
	01/01/2006 - 12/31/2006	56	44	468	346
	01/01/2007 - 12/31/2007	64	47	755	542
	01/01/2008 - 12/31/2008	83	68	1673	985
	01/01/2009 - 12/31/2009	97	83	758	550
Total:		362	288	5289	3212
NC7-sun.wilds	01/01/2005 - 12/31/2005	53	38	1056	783
	01/01/2006 - 12/31/2006	45	40	1072	648
	01/01/2007 - 12/31/2007	42	38	1302	1106
	01/01/2008 - 12/31/2008	58	52	1059	754
	01/01/2009 - 12/31/2009	67	60	822	572
Total:		265	228	5311	3863
Marek Total:		1521	1295	29778	19879

Millard

NC7-corn.kin	01/01/2005 - 12/31/2005	7	7	11	6
	01/01/2006 - 12/31/2006	16	14	34	6
	01/01/2007 - 12/31/2007	14	14	28	6
	01/01/2008 - 12/31/2008	15	15	25	6
	01/01/2009 - 12/31/2009	10	10	21	7
Total:		62	60	119	31
NC7-maize.gems	01/01/2005 - 12/31/2005	13	12	142	66
	01/01/2006 - 12/31/2006	28	25	334	66
	01/01/2007 - 12/31/2007	23	22	381	67
	01/01/2008 - 12/31/2008	27	23	329	81
	01/01/2009 - 12/31/2009	25	18	324	87
Total:		116	100	1510	367
NC7-maize.inb	01/01/2005 - 12/31/2005	161	132	1633	604
	01/01/2006 - 12/31/2006	265	197	2956	760
	01/01/2007 - 12/31/2007	259	203	3314	919
	01/01/2008 - 12/31/2008	271	210	5216	1634
	01/01/2009 - 12/31/2009	310	237	3332	916
Total:		1266	979	16451	4833
NC7-maize.pop	01/01/2005 - 12/31/2005	149	135	1302	1005
	01/01/2006 - 12/31/2006	195	163	2132	1463
	01/01/2007 - 12/31/2007	207	174	1722	1016
	01/01/2008 - 12/31/2008	234	191	2338	1547
	01/01/2009 - 12/31/2009	321	278	3930	2810
Total:		1106	941	11424	7841
NC7-maize.pvp	01/01/2005 - 12/31/2005	103	51	1095	76
	01/01/2006 - 12/31/2006	214	95	2202	111
	01/01/2007 - 12/31/2007	188	106	3181	130
	01/01/2008 - 12/31/2008	190	107	2340	153
	01/01/2009 - 12/31/2009	235	124	3729	194
Total:		930	483	12547	664

	NC7-maize.wilds	01/01/2005 - 12/31/2005	46	43	253	77
		01/01/2006 - 12/31/2006	59	49	303	77
		01/01/2007 - 12/31/2007	67	62	272	43
		01/01/2008 - 12/31/2008	60	58	201	42
		01/01/2009 - 12/31/2009	64	55	313	149
	Total:		296	267	1342	388
	Zea.totals	01/01/2005 - 12/31/2005	381	275	4425	1828
		01/01/2006 - 12/31/2006	585	356	7927	2477
		01/01/2007 - 12/31/2007	553	376	8870	2175
		01/01/2008 - 12/31/2008	601	406	10424	3457
		01/01/2009 - 12/31/2009	768	522	11628	4156
	Total:		2888	1935	43274	14093
	Millard Total:		2950	1995	43393	14124
Qu	NC7-medicinals	01/01/2005 - 12/31/2005	58	49	378	185
		01/01/2006 - 12/31/2006	44	35	323	163
		01/01/2007 - 12/31/2007	58	48	358	181
		01/01/2008 - 12/31/2008	34	31	232	161
		01/01/2009 - 12/31/2009	64	52	267	154
	Qu Total:		258	215	1558	844
Reitsma	NC7-chicory	01/01/2005 - 12/31/2005	9	9	257	118
		01/01/2006 - 12/31/2006	10	9	44	38
		01/01/2007 - 12/31/2007	5	5	203	162
		01/01/2008 - 12/31/2008	13	11	230	146
		01/01/2009 - 12/31/2009	19	18	35	26
	Total:		56	52	769	490
	NC7-cucumis	01/01/2005 - 12/31/2005	102	91	4761	2140
		01/01/2006 - 12/31/2006	126	108	3970	2096
		01/01/2007 - 12/31/2007	115	94	3368	1866
		01/01/2008 - 12/31/2008	121	106	3583	2033
		01/01/2009 - 12/31/2009	213	193	3854	2444
	Total:		677	592	19536	10579
	NC7-cucurbita	01/01/2005 - 12/31/2005	51	49	1567	828
		01/01/2006 - 12/31/2006	58	52	424	300
		01/01/2007 - 12/31/2007	41	36	525	323
		01/01/2008 - 12/31/2008	71	65	436	248
		01/01/2009 - 12/31/2009	114	108	1571	817
	Total:		335	310	4523	2516
	NC7-daucus	01/01/2005 - 12/31/2005	24	24	493	375
		01/01/2006 - 12/31/2006	33	32	460	356
		01/01/2007 - 12/31/2007	24	21	330	284
		01/01/2008 - 12/31/2008	29	26	684	475
		01/01/2009 - 12/31/2009	83	79	802	491
	Total:		193	182	2769	1981
	NC7-ocimum	01/01/2005 - 12/31/2005	23	21	204	85
		01/01/2006 - 12/31/2006	21	21	123	69
		01/01/2007 - 12/31/2007	11	10	68	51
		01/01/2008 - 12/31/2008	20	19	137	91
		01/01/2009 - 12/31/2009	39	37	442	91
	Total:		114	108	974	387
	NC7-parsnips	01/01/2005 - 12/31/2005	1	1	1	1
		01/01/2006 - 12/31/2006	3	3	22	15
		01/01/2007 - 12/31/2007	3	3	8	8
		01/01/2008 - 12/31/2008	2	2	6	6
		01/01/2009 - 12/31/2009	11	11	80	46
	Total:		20	20	117	76

Widrechner	Reitsma Total:		1395	1264	28688	16029
	NC7-mints	01/01/2005 - 12/31/2005	18	17	61	38
		01/01/2006 - 12/31/2006	19	19	55	37
		01/01/2007 - 12/31/2007	10	10	54	47
		01/01/2008 - 12/31/2008	14	14	88	64
		01/01/2009 - 12/31/2009	33	31	179	92
	Total:		94	91	437	278
	NC7-ornamentals	01/01/2005 - 12/31/2005	67	61	253	186
		01/01/2006 - 12/31/2006	87	74	428	316
		01/01/2007 - 12/31/2007	75	71	265	194
		01/01/2008 - 12/31/2008	88	82	342	246
		01/01/2009 - 12/31/2009	110	95	607	390
	Total:		427	383	1895	1332
	Widrechner Total:		521	474	2332	1610
<hr/>						
NCRPIS Total:		7901	6410	122539	62613	

Table 6A NC7 NIFA Regional Order History

TIME PERIOD	Total Number of Orders	Number of Orders (DI)	Foreign Orders (DI)		Domestic Orders (DI)		Domestic Orders (DI) NIFA Regions		
			Orders (DI)	Orders (DI)	Orders (DI)	Orders (DI)	NC7	NE9	S9
01/01/2009 to 12/31/2009	1823	1487	227	1260	601	134	290	235	
01/01/2008 to 12/31/2008	1579	1239	233	1006	486	113	201	206	
01/01/2007 to 12/31/2007	1491	1138	240	898	455	113	181	149	
01/01/2006 to 12/31/2006	1507	1182	249	933	506	110	184	133	
01/01/2005 to 12/31/2005	1225	930	175	755	348	77	198	132	
01/01/2004 to 12/31/2004	1045	787	164	623	287	71	159	106	
01/01/2003 to 12/31/2003	869	613	158	455	204	49	99	103	

Note: Total Number of Orders includes all orders handled, including NC7 orders.

Table 6B NC7 NIFA Regional Packet History

TIME PERIOD	Total Number of Packets	Number of Packets (DI)	Foreign Packets (DI)	Domestic Packets (DI)	Domestic Packets (in DI orders)			
					NC7	NE9	S9	W6
01/01/2009 to 12/31/2009	36754	26904	5440	21464	10921	2618	4096	3829
01/01/2008 to 12/31/2008	40682	24830	6719	18111	9268	1736	3561	3546
01/01/2007 to 12/31/2007	39066	22258	7766	14492	7218	1230	3433	2611
01/01/2006 to 12/31/2006	36383	26080	11950	14130	7483	1551	2354	2742
01/01/2005 to 12/31/2005	34111	22474	7510	14964	4434	2416	4723	3391
01/01/2004 to 12/31/2004	27225	17404	5539	11865	4237	3449	1823	2356
01/01/2003 to 12/31/2003	24327	11855	4666	7189	4230	508	1205	1246

Note: Total Number of Packets includes all packets handled, including NC7 distributed.

Figure 1

