2006 POTATO BREEDING AND GENETICS RESEARCH REPORT

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INTRODUCTION

At Michigan State University we are breeding potatoes for the chip-processing and tablestock markets. The program is one of four integrated breeding program in the North Central region. At MSU, we conduct a multi-disciplinary program for potato breeding and variety development that integrates traditional and biotechnological approaches. In Michigan, it requires that we primarily develop high yielding round white potatoes with excellent chip-processing from the field and/or storage. We conduct variety trials of advanced selections and field experiments at MSU research locations (Montcalm Research Farm, Lake City Experiment Station, Muck Soils Research Farm and MSU Soils Farm), we ship seed to other states and Canadian provinces for variety trials, and we cooperate with Chris Long on 23 grower trials throughout Michigan. Through conventional crosses in the greenhouse, we develop new genetic combinations in the breeding program, and also screen and identify exotic germplasm that will enhance the varietal breeding efforts. With each cycle of crossing and selection we are seeing directed improvement towards improved varieties (e.g. combining chip-processing, scab resistance and late blight resistance). In addition, our program has been utilizing genetic engineering as a tool to introduce new genes to improve varieties and advanced germplasm for traits such as solids, insect resistance, disease resistance and nutritional enhancement. We feel that these in-house capacities (both conventional and biotechnological) put us in a unique position to respond to and focus on the most promising directions for variety development and effectively integrate the breeding of improved chip-processing and tablestock potatoes.

The breeding goals at MSU are based upon current and future needs of the Michigan potato industry. Traits of importance include yield potential, disease resistance (scab, late blight, early die and PVY), insect (Colorado potato beetle) resistance, chipping (out-of-the-field, storage, and extended cold storage) and cooking quality, bruise resistance, storability, along with shape, internal quality and appearance. We are also developing potato tuber moth resistant lines as a component of our international research project. If these goals can be met, we will be able to reduce the grower's reliance on chemical inputs such as insecticides, fungicides and sprout inhibitors, and improve overall agronomic performance with new potato varieties.

Over the years, key infrastructure changes have been established for the breeding program to make sound assessments of the breeding selections moving through the program. These include the establishment and expansion of the scab nursery, the development of the Muck Soils Research Farm for late blight testing, the incorporation of no-choice caged studies for Colorado potato beetle assessment, the Michigan Potato Industry Commission (MPIC)-funded construction of the B.F. (Burt) Cargill Demonstration Storage adjacent to the Montcalm Research Farm, new land at the Lake City Experiment Station along with a well for irrigation and expanded land at the Montcalm Research Farm.

PROCEDURE

I. Varietal Development

Each year, during the winter months, 500-1000 crosses are made using about 150 of the most promising cultivars and advanced breeding lines. The parents are chosen on the basis of yield potential, tuber shape and appearance, chip quality, specific gravity, disease resistance, adaptation, lack of internal and external defects, etc. These seeds are then used as the breeding base for the program. We also obtain seedling tubers or crosses from other breeding programs in the US. The seedlings are grown annually for visual evaluation (size, shape, set, internal defects) at the Montcalm and Lake City Research Farms as part of the first year selection process of this germplasm each fall. Each selection is then evaluated post harvest for specific gravity and chip processing. These selections each represent a potential variety. This system of generating new seedlings is the initial step in an 8-12 year process to develop new varieties. This step is followed by evaluation and selection at the 8-hill, 20-hill and 30-hill stages. The best selections out of the four-year process are then advanced for testing in replicated trials (Preliminary, Adaptation, Dates-of-Harvest, Grower-cooperator trials, North Central Regional Trials, Snack Food Association Trials, and other out-of-state trials) over time and locations. The agronomic evaluation of the advanced breeding lines in the replicated trials is reported in the annual Potato Variety Evaluation Report.

II. Evaluation of Advanced Selections for Extended Storage

With the Demonstration Storage facility adjacent to the Montcalm Research Farm we are positioned to evaluate advanced selections from the breeding program for chip-processing over the whole extended storage season (October-June). Tuber samples of our elite chip-processing selections are placed in the demonstration storage facility in October and are sampled monthly to determine their ability to chip-process from colder (42-48°F) and/or 50˚F storage. In addition, Chris Long evaluates the more advanced selections in the 10 cwt box bins and manages the 500 cwt. storage bins which may have MSU-bred lines. In 2006-7 we are incorporating a preliminary sugar profiling evaluation of advanced selections from the breeding program.

III. Germplasm Enhancement

To supplement the genetic base of the varietal breeding program, we have a "diploid" (2x = 24 chromosomes) breeding program in an effort to simplify the genetic system in potato (which normally has 4x chromosomes) and exploit more efficient selection of desirable traits. This added approach to breeding represents a large source of valuable germplasm, which can broaden the genetic base of the cultivated potato. The diploid breeding program germplasm base at MSU is a synthesis of seven species: *S. tuberosum* (adaptation, tuber appearance), *S. raphanifolium* (cold chipping), *S. phureja* (cold-chipping, specific gravity, PVY resistance, self-compatability), *S. tarijense* and *S. berthaultii* (tuber appearance, insect resistance, late blight resistance, verticillium wilt resistance), *S. microdontum* (late blight resistance) and *S. chacoense* (specific gravity, low sugars, dormancy and leptine-based insect resistance). In general, diploid breeding utilizes haploids (half the chromosomes) from potato varieties, and diploid wild and cultivated tuber-bearing relatives of the potato. Even though these potatoes have only half the chromosomes of the varieties in the U.S., we can cross these potatoes to transfer the desirable genes by conventional crossing methods via 2n pollen.

IV. Integration of Genetic Engineering with Potato Breeding

Through transgenic approaches we have the opportunity to introduce new genes into our cultivated germplasm that otherwise would not be exploited. It has been used in potato as a tool to improve commercially acceptable cultivars for specific traits. Our laboratory has 14 years experience in *Agrobacterium*-mediated transformation to introduce genes into important potato cultivars and advanced breeding lines. We are presently using genes in vector constructs that confer resistance to Colorado potato beetle and potato tuber moth (*Bt-cry3A*, *Bt-cry1Ia1* and avidin), potato tuber moth, late blight resistance via the *RB* gene, drought resistance (*CBF1*) and vitamin E. Furthermore, we are investing our efforts in developing new vector constructs that use alternative selectable markers and give us the freedom to operate from an intellectual property rights perspective. In addition, we are exploring transformation techniques that eliminate the need for a selectable marker (antibiotic resistance) from the production of transgenic plants.

RESULTS AND DISCUSSION

I. Varietal Development

Breeding

The MSU potato breeding and genetics program is actively producing new germplasm and advanced seedlings that are improved for cold chipping, and resistance to scab, late blight, and Colorado potato beetle. For the 2006 field season, progeny from over 500 crosses were planted and evaluated. Of those, the majority were crosses to select for round whites (chip-processing and tablestock), with the remainder to select for yellow flesh, long/russet types, red-skin, and novelty market classes. In addition to crosses from the MSU breeding program, crosses were planted and evaluated from collaborative germplasm exchange from other breeding programs including North Dakota State University, University of Minnesota, and the USDA/ARS program at the University of Wisconsin as part of the Quad state cooperative effort. During the 2006 harvest, over 800 selections were made from the 40,000 seedlings produced. All potential chip-processing selections will be tested in January or March 2007 directly out of 40°F and 45°F storages. Atlantic (50°F chipper) and Snowden (45°F chipper) are chip-processed as check cultivars. Selections have been identified at each stage of the selection process that have desirable agronomic characteristics and chip-processing potential. At the 8-hill and 20-hill evaluation state, over 400 and 100 selections were made, respectively. Selection in the early generation stages has been enhanced by the incorporation of the Colorado potato beetle, scab and late blight evaluations of the early generation material.

Chip-Processing

Over 80% of the single hill selections have a chip-processing parent in their pedigree. Based upon the pedigrees of the parents we have identified for breeding cold-chipping potato varieties, there is a diverse genetic base. We have at least eight cultivated sources of cold-chipping. Examination of pedigrees shows up to three different cold-chipping germplasm sources have been combined in these selections. Our promising chip-processing lines are MSJ147-1, MSJ036-A (scab resistant), MSH228-6 (moderate scab resistance), MSJ126-9Y (moderate scab resistance), MSJ316-A (moderate scab resistance), MSK061-4 (moderate scab resistance), MSK409-1 (scab resistant), MSN099-B (scab resistance), MSN238-A (scab resistance), MSL007-B (scab resistance) and MSQ070-1 (scab and late blight resistant).

Dr. Joe Sowokinos, Univ. of Minnesota, has conducted biochemical analyses of our best chipping lines and has discovered that our lines differ from older varieties in their proteins (UGPase) involved in chipping. Some of these lines are MSJ147-1, MSG227-2 and MSJ126-9Y. Moreover, MSJ147-1 and MSJ126-9Y have the desirable levels of acid invertase to chip process from colder storage. We are using this information to help us design specific crosses to find improved chip-processing varieties that will allow processing from colder storage temperatures.

Tablestock

Efforts have been made to identify lines with good appearance, low internal defects, good cooking quality, high marketable yield and resistance to scab and late blight. Our current tablestock development goals now are to continue to improve the frequency of scab resistant lines, incorporate resistance to late blight along with marketable maturity and excellent tuber quality, and select more russet and yellow-fleshed lines. From our breeding efforts we have identified mostly round white lines, but we also have a number of yellow-fleshed and red-skinned lines, as well as long, russet type and purple skin selections that carry many of the characteristics mentioned above. We are also selecting for a dual-purpose russet, round white, red-skin, and improved Yukon Gold-type yellow-fleshed potatoes. Some of the tablestock lines were tested in on-farm trials in 2006, while others were tested under replicated conditions at the Montcalm Research Farm. Promising tablestock lines include MSE221-1 as a scab resistant tablestock, and MSN084-3, a round white with a smooth round shape and bright skin. We have a number of tablestock selections with late blight resistance. These are MSL072-C and MSM171-A. MSL211-3 and MSN105-1 has late blight and scab resistance. MSE192-8RUS and MSA8254-2BRUS are two russet table selections that have scab resistance, while MSL794-BRUS has late blight resistance.. MSI005-20Y is a yellow-fleshed line with smooth round appearance and high yield potential. Some newer lines with promise include MSQ176-5, MSQ441-6R, MSN230-6RY and MSQ087-3.

Disease and Insect Resistance Breeding

Scab: Disease screening for scab has been an on-going process since 1988. Results from the 2006 MSU scab nursery indicate that 21 of 176 lines evaluated had a scab rating of 1.0

or less (little to no infection to common scab). The limitation of breeding for scab resistance is the reliance on the scab nursery. The environmental conditions can influence the infection each year, thus multiple year data provides more reliable data. A study was conducted to develop a laboratory-based screening process that would use thaxtomin in tissue culture and tuber disks to expedite selection of material with potential scab resistance. We found no correlation between thaxtomin reaction and scab rating. In addition, we found a moderate correlation between the field screening and greenhouse screening for scab. In 2004, we expanded the scab nursery with an additional acre of land nearby. This expansion has allowed us to conduct early generation selection for scab resistance among our breeding material. In 2006, 110 of 293 early generation selections showed strong scab resistance. These data were incorporated into the early generation evaluation process at Lake City. We are seeing that this expanded effort is leading to more scab resistant lines advancing through the breeding program.

Late Blight: With support from GREEEN, the Muck Soils Research Farm, Bath, Michigan has become an excellent North American site for late blight testing because of the humid microclimate and isolation from major commercial potato production. As a result, late blight infection has been consistently achieved each year making breeding efforts to select late blight resistant germplasm very efficient. In 2006 untimely flooding led to the loss of the field experiments. No data was collected.

Colorado potato beetle: With support from GREEEN, we also introduced an early generation Colorado potato beetle screen at the Montcalm Research Farm. In 2006, 480 clones from 52 families were evaluated at the Montcalm Research Farm Beetle Nursery. The beetle pressure was extremely high leading to complete defoliation in all susceptible check lines. Percent defoliation was visually estimated during the beetle infestation in June and July. The lines were then sorted into four categories: susceptible, reduced susceptibility, moderately resistant and resistant. From 15 families, 72 clones were classified as resistant. The majority of the lines that were moderately resistant or resistant can be attributed to the expression of the *Bt-cry3A* gene or glycoalkaloid/leptine based mechanisms. The most resistant material was selected for further advancement in the breeding program and also for use in the next round of crossing to develop beetle resistant cultivars. Concurrently, a field cage (no-choice) experiment was conducted to evaluate 6 lines. In 2006 beetle behavior was evaluated in lines that expressed differing levels of Bt genes. The data from this experiment has not been analyzed yet.

It is a great challenge to achieve host plant resistance in a commercially acceptable line. We have some promising advanced selections with partial resistance to Colorado potato beetle. In addition, we have *Bt-cry3A* transgenic lines that could be commercialized if the processors renewed their acceptance and regulatory environment was modified to reduce costs. I am on a national committee to help build infrastructure to so that transgenic specialty crops like potato can be deregulated in a more efficient and less costly manner.

II. Evaluation of Advanced Selections for Extended Storage: MSU Potato Breeding Chip-processing Results From the MPIC Demonstration Commercial Storage (October 2005 - June 2006)

The MSU Potato Breeding Program has been conducting chip-processing evaluations each year on potato lines from the MSU breeding program and from other states. For 7 years we have been conducting a long-term storage study to evaluate advanced breeding lines with chip-processing potential in the Dr. B. F. (Burt) Cargill Potato Demonstration Storage facility directly adjacent to the MSU Montcalm Research Farm to identify extended storage chippers. We evaluated advanced selections from the breeding program for chip-processing over the whole extended storage season (October-June). Tuber samples of our elite chip-processing selections are placed in the demonstration storage facility in October and are sampled monthly to determine their ability to chip-process from storage. In addition, Chris Long and the Storage Committee evaluate the more advanced selections in the 10 cwt box bins and manage the 500 cwt. storage bins which may include MSU lines.

In October 2005, tuber samples from 5 MSU lines, 2 Frito Lay lines, Beacon Chipper, NY132, along with the standards of Pike and Atlantic from the Montcalm Research Farm trials were placed in the bin to be cooled to 48°F. The bin temperature in December was 52°F and reached a low of 48°F in February. Tubers from 5 other MSU lines, FL1879, USDA line A91814-2, W2128-8 and Snowden were placed in the bin that was to be cooled to 46°F. The bin was at 50°F in November and down to 46°F by January. The first samples were chip-processed at MSU in November and then, each month until May 2005. Samples were evaluated for chip-processing color and quality.

Table 1 summarizes the chip-processing color of select lines over the 7-month storage season. In the 48°F temperature bin, Atlantic and Pike were the check varieties. From November to March all lines chip-processed acceptably. This is not surprising since the best chip-processing lines were selected for this study. In some cases, SED or hollow heart was observed in a few chips, but no patterns emerged. The storage test was terminated in mid-May. Based upon the data, many of these lines have potential to be further tested in storage tests. Based upon chip color and defects the most promising chip-processing lines for storage and scab resistance are Beacon Chipper, MSJ036-A, MSH228-6, NY132 and FL1922. MSJ461-1 also has strong foliar resistance to late blight.

In the bin for colder temperature storage (46°F), Snowden was used as check variety and chip-processed acceptably except in May. All other lines produced acceptable chips throughout the storage season. MSG227-2 and MSK061-4, MSM051-3 offer scab resistance along with their ability to chip-process. MSJ147-1 was one line that gave consistently low color scores through out the season. See Chris Long’s storage report for those results and results from the box bins and 500cwt storage bins.

III. Germplasm Enhancement

In 2006, less than 3% of the populations evaluated as single hills were diploid. From this breeding cycle, we plan to screen the selections chip-processing from storage. In addition, selections were made from over progeny that was obtained from the USDA/ARS at the University of Wisconsin. These families represent material from South American potato species and other countries around the world that are potential sources of resistance to Colorado potato beetle, late blight, potato early die, and ability to cold-chip process. Through GREEEN funding, we were able to initiate a breeding effort to introgress leptine-based insect resistance. From previous research we determined that the leptine-based resistance is effective against Colorado potato beetle. We will continue conducting extensive field screening for resistance to Colorado potato beetle at the Montcalm Research Farm and at the Michigan State University Horticulture Farm in 2007. In 2004 we made crosses with late blight resistant diploid lines derived from *Solanum microdontum* to our tetraploid lines. This *S. microdontum*-based resistance is unique and very effective against the US-8 strains. These progeny are being grown in the greenhouse and now we have used DNA marker analysis to identify which lines have the late blight resistance. We have lab-based detached leaf bioassays planned for the winter of 2007 to evaluate these lines.

IV. Integration of Genetic Engineering with Potato Breeding

Combining engineered and natural host plant resistance to *Phytophthora infestans* in cultivated potato

General susceptibility of potato cultivars to *Phytophthora infestans* (Mont.) de Bary is a major concern for potato production. The major resistance gene *RB* was cloned from *Solanum bulbocastanum* Dun.a diploid (2n=2x=24) Mexican species that is highly resistant to all known races of *P. infestans*. The objective of this work is to combine conventionally bred sources of resistance with the *RB* gene via *Agrobacterium* transformation. Our hypothesis is that by pyramiding engineered resistance with natural plant resistance we expect to obtain stronger and more durable resistance to potato late blight. Therefore, this study was undertaken to test the effectiveness of the *RB* gene on its own by transforming late blight-susceptible clones (Spunta, and the breeding lines MSE149-5Y and MSG227-2), and to test the effectiveness of the gene in combination with natural late blight resistance by transforming resistant clones (Stirling, and the advanced breeding line MSJ461-1). In 2005 we identified 5 lines with RB-based late blight resistance (MSE149-5Y, Spunta and MSG227-2) at the Muck Soils Research Farm trials. No data collected in 2006. The Spunta and MSE149-5Y lines were used in crosses to transfer the RB-based resistance to other genetic backgrounds in the breeding program. We have conducted tissue culture regeneration studies this past year to identify the best lines we can use in our RB transformation studies. In addition, we have created new vectors that may help us achieve high transformation efficiency with the RB gene.

Insecticidal activity of avidin against Colorado potato beetle larvae, *Leptinotarsa decemlineata* (Say)

The Colorado potato beetle, *Leptinotarsa decemlineata* (Say), is the most destructive insect pest of potato, *S. tuberosum* (L.) in eastern North America. Biotin is an essential co-enzyme required for all organisms, including insects. Avidin binds to biotin, therefore limiting its availability during insect growth and development. Without this co-enzyme, an insect's growth is severely stunted, eventually leading to death. We have expressed avidin in two potato lines: MSE149-5Y, a susceptible potato line, and ND5873-15, a high glycoalkaloid line. The expression levels of avidin in the transgenic MSE149-5Y and ND5873-15 was determined to be 108.6 ± 0.4μg /mg and 108.2± 0.9μg/mg, respectively. Detached leaf bioassays were performed on transgenic and non-transgenic lines of MSE149-5Y and ND5873-15 using Colorado potato beetle first and third stage larvae. First stage larvae survivorship was significantly less for larvae feeding transgenic MSE149-5Y and ND5873-15 lines compared to the non-transgenic lines, but third stage larvae survivorship did not significantly differ for larvae feeding on transgenic MSE149-5Y and ND5873-15 lines compared to larvae feeding on non-transgenic lines. The growth of first stage larvae was significantly stunted for larvae feeding on the transgenic MSE149-5Y and ND5873-15 lines compared to the non-transgenic lines. Subsequently, we placed neonates on non-transgenic and transgenic MSE149-5Y and ND5873-15 lines and monitored growth and development of insects. Avidin expressing potato plants appearred to delay development and resulted in significantly less emerging adults. Potatoes expressing avidin may have value in managing insect pests.

Commercialization of Potato Tuber Moth Resistant Potatoes in South Africa

The potato tuber moth (*Phthorimaea operculella* Zeller) is a primary insect problem facing potato farmers in developing countries. Currently, the only available means to control the potato tuber moth (PTM) and avoid major crop losses is the use of chemical pesticides, however, it is problematic to use this strategy to control the tuberworm in storage potatoes. Michigan State University (MSU), funded by the U.S. Agency for International Development (USAID) through its Agricultural Biotechnology Support Project (ABSP), initiated biotechnology research on the development of PTM resistant varieties in 1992. A *Bacillus thuringiensis* (Bt)-*cry1Ia1* gene, was obtained from ICI Seeds (now Syngenta) and successfully introduced into several potato varieties, including Spunta. Transgenic lines and their progeny were shown to have excellent control of the larvae. The Bt Potato will be the first public sector-developed products to reach farmers in developing countries and will serve as a model for the public-sector deployment of crops that are resistant to insects. When the farmers choose to grow the Bt potato, the benefits to the farmer and end-users will be reduced input costs (less insecticides used), increased marketable yield, improved quality, reduced post-harvest losses, reduced human exposure to pesticides, and less pesticide residues on potato tubers. The commercialization project includes six components: Product Development, Regulatory File Development, Obtaining Freedom to Operate and Establishing Licensing Relationships, Marketing and Technology Delivery, Documentation of Socio-Economic Benefits, and Public Communication. This technology would also have benefits in controlling PTM in the US and reducing the need for insectcide-based protection.

V. Variety Release

No lines are planned for release in 2007, but we are continuing to promote the seed production and testing of Beacon Chipper, a 2005 release. In addition, we are continuing to promote Michigan Purple, Jacqueline Lee, MSE192-8-RUS, A8254-2BRUS, MSI005-20Y and MSN105-1 for the tablestock markets. Lastly, commercial seed of MSH228-6, MSJ147-1, MSK061-4, MSJ126-9Y and MSJ036-A are being produced and we will continue to seek commercial testing of these lines. We have also initiated a focused ribavirin-based virus eradication system to generate virus-free lines for the industry.

2005-2006 Demonstration Storage Chip Results
Michigan State University Potato Breeding and Genetics
Montcalm Research Farm
Chip Scores: SFA Scale†

		Sample Dates:					
	Date:	11/8/2005	12/14/2005	1/10/2006	2/8/2006	3/8/2006	5/15/2006
Line	**Temp:**	50 °F	52 °F	48 °F	48 °F	52 °F	53 °F
ATLANTIC		**1.5**	**1.5**	**1.5**	**1.0**	**1.0**	**1.5**
Beacon Chipper		1.0	1.0	1.0	1.0	1.0	1.5
FL1833		1.0	1.0	1.0	1.0	1.0	1.0
FL1922		1.5	1.0	1.0	1.0	1.0	1.0
MSH228-6		1.0	1.0	1.0	1.0	1.0	-
MSJ036-A		1.5	1.0	1.5	1.0	1.0	2.0
MSJ316-A		1.0	1.5	1.0	1.0	1.0	1.0
MSJ461-1		1.5	1.5	1.5	1.0	1.0	1.0
MSK049-A		1.5	1.0	1.0	1.0	1.0	-
NY132		1.0	1.0	1.0	1.0	1.5	1.0
PIKE		**1.0**	**1.0**	**1.0**	**1.0**	**1.0**	**1.0**
	Temp:	50 °F	46 °F	46 °F	46 °F	46 °F	53 °F
A91814-2		1.5	1.0	1.5	1.0	1.0	1.5
FL1879		1.5	1.0	1.0	1.0	1.0	1.0
MSG227-2		1.0	1.0	1.0	1.0	1.0	1.0
MSJ147-1		1.0	1.0	1.0	1.0	1.0	1.5
MSK009-B		1.0	1.0	1.0	1.0	1.0	2.0
MSK061-4		1.0	1.0	1.0	1.0	1.0	1.5
MSM051-3		1.0	1.0	1.0	1.0	1.0	1.5
SNOWDEN		**1.0**	**1.0**	**1.0**	**1.0**	**1.0**	**2.5**
W2128-8		1.5	1.0	1.5	1.0	1.0	1.5

†Snack Food Association Chip Score

Ratings: 1 - 5
1: Excellent
5: Poor

Chip scores were from two-slice samples from five tubers of each line collected at each sample date.