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Only a few persons have the wisdom to understand state, regional and national needs in their fields. Dr. Richard Chase is one of them. No other single person has done so much for the potato industry. His vision and attention to detail have helped advance the industry's research and education programs not only in Michigan but nationwide. Our thanks for his lifetime of accomplishments, for the influence he has had on so many others during his life's journey, expresses itself in many forms. This publication offers a fitting opportunity to demonstrate our gratitude once more. I am pleased to dedicate the 2002 Michigan Potato Industry Commission Research Report to Dr. Richard Chase.

Don Sklarczyk  
Research Committee Chairman

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# 2002 POTATO BREEDING AND GENETICS RESEARCH REPORT

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## INTRODUCTION

At MSU, we conduct a multi-disciplinary program for potato breeding and variety development that integrates traditional and biotechnological approaches. 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 16 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 and disease resistance. 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 and early die), 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.

## 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 and 20-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 (42-46°F) and/or 50°F storage.

## **III. Germplasm Enhancement**

To supplement the genetic base of the varietal breeding program, we have a "diploid" (2x 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-compatibility), *S. tarijense* and *S. berthaultii* (tuber appearance, insect 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 10 years experience in *Agrobacterium*-mediated transformation to introduce genes into important potato cultivars and advanced breeding lines. We presently have genes in vector

constructs that confer resistance to PVY, Colorado potato beetle, potato tuber moth, broad-spectrum disease resistance via the glucose oxidase (GO) gene, late blight resistance with the resveratrol synthase (RS) and divinyl ether synthase (DES) genes, and cold/frost resistance (COR15). We also have the *glgC16* gene (ADP-glucose pyrophosphorylase (AGPase) or starch gene) from Monsanto to modify starch and sugar levels in potato tubers. 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 selectable marker (antibiotic resistance) from the transgenic plants.

## V. Variety Release

Beginning in 2002, the MSU breeding program has named and released its first three varieties and is in the process of licensing these new varieties to the Michigan Potato Industry Commission. Each year the best lines will be considered for release. In 2003 MSF373-8, with the name Boulder being considered, will be brought forward.

## 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 2002 field season, progeny from over 600 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. During the 2002 harvest, about 1200 selections were made from the 35,000 seedlings grown at the Montcalm Research Farm. Following harvest, specific gravity was measured and potential chip-processing selections were chipped out of the field. All potential chip-processing selections will be tested in January or March 2003 directly out of 42°F and 50°F storage. Atlantic (50°F chipper) and Snowden (45°F chipper) are chipped 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, 450 and 140 selections were made, respectively. **Table 1** lists some of the potential lines for grower trials in year 2003.

#### Chip-Processing

Excellent chip-processing selections have been identified in the breeding program, despite switching to a more stringent screening temperature (42 vs. 45°F storage) a few years ago. Over 70% of the single hill selections have a chip-processing parent in their pedigree. Of those selections, about 75% have a SFA chip score of 1.5 or less. Based upon the pedigrees of the parents we have identified for breeding cold-

chipping potato varieties, we have a diverse genetic base. We believe that we have at least eight cultivated sources of cold-chipping. We have made various hybrid combinations with these parents from which to pyramid cold-chipping traits and the hybrid populations have been grown out, selected and evaluated. We now have advanced into the crossing block these new MSU selections that have chip quality directly from 42°F storage. Examination of pedigrees shows up to three different cold-chipping germplasm sources have been combined in these selections. Promising chip-processing lines are MSF099-3 (42°F chipper), MSG227-2 (scab resistant 45°F chipper), MSH095-4, MSH094-8, MSH067-3, MSJ147-1, MSJ126-9Y, MSJ167-1, and late blight resistant chipper MSJ461-1.

### **Tablestock**

One of our objectives is also to develop improved cultivars for the tablestock industry. 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. From our 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 2002, while others were tested under replicated conditions at the Montcalm Research Farm. Promising tablestock lines include MSE221-1 as a scab resistant tablestock, while MSE018-1 is a high yielding tablestock with a large oval shape. MSE192-8RUS and MSE202-3RUS are two russet table selections that have excellent type and scab resistance. MSE149-5Y, MSI005-20Y and MSJ033-6Y are yellow-fleshed lines with smooth round appearance and high yield potential. MSF373-8 is a high yielding line with large tubers that also chip out of the field. This line is being considered for release as Boulder. 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 lines.

### **Disease and Insect Resistance Breeding**

Disease screening for scab has been an on-going process this 1988. Results from the 2002 MSU scab nursery indicate that 16 of 160 lines evaluated demonstrated strong resistance (no evidence of infection) to common scab in 2002. In addition, 10 other MSU breeding lines showed moderate scab resistance. 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 laboratory-based screening process is currently under development that would use thaxtomin in tissue culture to expedite selection of material with potential scab resistance.

Since the mid-1990's we have directed efforts to identify sources of late blight resistance and use this resistance to breed late blight resistant varieties. At MSU, we have also participated in the national late blight trial and we have conducted our own efforts to use field and greenhouse screening to identify additional sources of resistance that can be used by the breeding community. In the past six years the MSU breeding



program has intensely evaluated over 700 crosses between late blight resistant x late blight susceptible parents and have identified parents that transmit strong late blight resistance to the highest percentage of the offspring. As of 2002, based upon six years of inoculated field experiments, we have at least eight sources of foliar resistance to the US8 genotype of *Phytophthora infestans* (Mont.) that have different pedigrees from which their resistance is derived. The resistance in Jacqueline Lee has now held resistance for six years of testing. MSJ461-1, the chip-processing selection, has the same late blight resistance source as Jacqueline Lee. Our other promising late blight resistant lines that have been tested in replicated agronomic trials are MSJ317-1, MSI152-A, MSJ453-4Y, MSJ456-4 and MSL757-1 (see Potato Variety Evaluation Report for agronomic data). In each of these lines, the resistance is based on a single resistance source. If we rely on a single source of resistance, the varieties developed from this strategy may be overcome by *P. infestans* at some future date that we cannot predict. Therefore, the most effective breeding strategy is to combine resistance from different pedigrees to build a more durable resistance. Our efforts are now focusing on pyramiding the different resistance sources.

Single-hill selections in 2002 also had an exciting number of individuals with pedigrees for potential late blight, Colorado potato beetle or scab resistance or some combination of the three. Of the single hill selections, 40% of progeny have at least one late blight parent, 15% have a Colorado potato beetle resistant parent, and 15% have a scab resistant parent in its pedigree.

## **II. Evaluation of Advanced Selections for Extended Storage: MSU Potato Breeding Chip-processing Results From the MPIC Demonstration Commercial Storage (October 2001 - June 2002)**

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 three years we have been conducting a 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. In October 2001, tuber samples from seven lines in the Montcalm Research Farm trials were placed in the bin to be cooled to 46°F. Tubers from another seven lines were placed in the bin that was to be cooled then held at 52°F. The first samples were chip-processed at MSU in October and then, each month until June 2002. Samples were evaluated for chip-processing color and quality.

**Table 2** summarizes the chip-processing color of select lines over the 8-month storage season. In the 46°F bin, Snowden was the check variety. In May the Snowden chips went off-color. In contrast only MSG227-2, MSH094-8 and MSH095-4 maintained acceptable chip color until the June 2002 sampling. Of these lines, MSG227-2 and MSH094-8 maintained the lightest chip color throughout the storage season. Chip-processing ability of MSG227-2 and NY112 was also observed during the past two year's storage studies in the Demonstration Storage Facility. MSG227-2 also has scab resistance. If the agronomic performance of MSG227-2 is considered acceptable, it will be considered for commercial release after the 2003 season.

In the 52°F bin Atlantic and Pike were used as check varieties and both varieties chip-processed acceptably until April. Of the six advanced breeding lines evaluated, Liberator and MSJ461-1 chip-processed acceptably throughout the storage season. Liberator offers chip-processing from storage and scab resistance. MSJ461-1 had the most consistent and lightest chip color throughout the storage season. MSJ461-1 also offers strong foliar late blight resistance along with the chip-processing quality; however the solids content is lower than other chip-processing lines.

In addition, MSF099-3 was grown by Sandyland Farms in 2001 and placed in one of the 500 cwt bins. Despite field frost occurring in the harvested tubers, the potatoes chip-processed successfully out of the bin in April 2002 at Utz in Pennsylvania.

### **III. Germplasm Enhancement**

In 2002, about 5% 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 3,000 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. About 100 selections were made among the diploid material in 2002. Through GREEN 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 be conducting extensive field screening for resistance to Colorado potato beetle in 2003.

### **Late Blight Breeding and Genetics: Mapping Late Blight Resistance in three Populations**

A high priority objective of the breeding program is to identify sources of late blight resistance and use these sources for breeding varieties with late blight resistance. In 1999 we initiated a set of studies (via GREEN) to identify the genes in potato associated with late blight resistance. If we can identify the genes that contribute to late blight resistance we feel that we could more effectively breed varieties with durable late blight resistance. A diploid potato population was developed with the objectives to map quantitative trait loci (QTL) conferring resistance to *Phytophthora infestans* (Mont.) de Bary and other agronomic traits using simple sequence repeats (SSR) and isozymes and to examine associations between late blight resistance and other agronomic traits. The mapping population was a cross between a late blight resistant selection of *Solanum microdontum* Bitter and a susceptible diploid advanced breeding clone. A second diploid population derives its late blight resistance from *S. berthaultii*. The third population is tetraploid and the resistance comes from Jacqueline Lee. Based upon field trials at the Muck Soils Research Farm, Bath, MI between 1999 and 2002, we have identified major late blight resistance genes in the three populations. Currently, one chromosome region containing the resistance is linked to a genetic marker has been identified in *S. microdontum*. Following the gene mapping analyses this winter, we will find the other two major resistances linking with a genetic The major QTL associated with late blight

resistance is suitable for marker-assisted selection to introgress a new source of resistance to *P. infestans* to the cultivated tetraploid germplasm of potato.

The tetraploid cross for mapping (Jacqueline Lee x MSG227-2) offers more than just mapping late blight resistance genes. This cross has traits such as late blight resistance, scab resistance, chip-processing, specific gravity, maturity all segregating at one time. The original cross had over 300 progeny. From those, about 75% had acceptable tuberization characteristics. Following late blight screening at the Muck Soils Research Farm, 41 progeny had foliar late blight resistance. Of those late blight resistant progeny, about 75% had acceptable yield and tuber type for selection. About 30 lines were chip-processed. Of those selections about 10 had acceptable chip-processing color, with 5 having acceptable solids levels in the tubers. About 25 of the selected progeny are being advanced for further evaluation in 2003.

#### **IV. Integration of Genetic Engineering with Potato Breeding**

The program has been conducting transformations of potato to introduce a variety of transgenes. Currently we have genetically engineered plants that express the *Bt-cry3A* gene to control the Colorado potato beetle, the glucose oxidase and resveratrol synthase genes for disease resistance, and the AGPase gene for low sugars and high solids.

#### **Assessment of Natural (Glandular Trichomes and Glycoalkaloid-Based) and Engineered (*Bt-cry3A*) Potato Host Plant Resistance Mechanisms for Control of Colorado potato beetle: Caged no-choice studies.**

The Colorado potato beetle, *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae), is the leading insect pest of potato (*Solanum tuberosum* L.) in northern latitudes. Host plant resistance is an important tool in an integrated pest management program for controlling insect pests. A field study was conducted in 2002 to compare natural (glandular trichomes (NYL235-4) and glycoalkaloid-based (ND5873-15)), engineered (*Bt-cry3A*: NO8.8), and combined (glandular trichomes + *Bt-cry3A* (L28.3) and glycoalkaloids + *Bt-cry3A* (ND8.01) transgenic potato lines) host plant resistance mechanisms of potato for control of Colorado potato beetle. Six different potato lines representing five different host plant resistance mechanisms were evaluated in caged studies (no-choice) at the MSU campus farms. Each cage with 10 plants represented one plot. The cages were arranged in a randomized complete block design consisting of three replications. Observations were recorded weekly for a visual estimation of percent defoliation by Colorado potato beetles, and the number of egg masses, larvae, and adults. The *Bt-cry3A* transgenic, and the combined resistance lines were effective in controlling feeding by Colorado potato beetle adults and larvae. Effectively no feeding was observed in the glycoalkaloid + *Bt-cry3A* transgenic line. The high glycoalkaloid line had less feeding, but the beetles clipped the petioles, which led to greater defoliation in the first few weeks. Foliage re-growth occurred by the end of the season. The glandular trichome line suffered less feeding than the susceptible control. Based on these results, the *Bt-cry3A* gene in combination with glandular trichome or glycoalkaloid-based host plant resistance mechanisms is an effective strategy that could be used to develop potato varieties for use in a resistance management program for control of Colorado potato beetle. **Figure 1** shows the results of caged trial in 2002.



### **Bt-cry3A-transgenic line Agronomic Trial**

In 2001 and 2002, we had extensive field testing for agronomic performance in replicated trials of our most advanced *Bt-cry3A* transgenic lines. Based upon 2001 agronomic performance and 2002 Bt-cry3A protein concentrations in foliage, 12 of 26 transgenic lines were eliminated. **Table 3** summarizes the results from the Advanced *Bt-cry3A* Breeding Line Preliminary Trial at the Montcalm Research Farm. In general, the *Bt-cry3A* transgenic lines had similar agronomic and tuber characteristics compared to the non-transgenic parental line. These selections represent a diverse portfolio of Bt-cry3A lines that could be commercialized if the intellectual property rights and regulatory requirements could be met. We will maintain these lines in our program. If the acceptance of transgenic food crops becomes deregulated, we will consider these lines for commercialization.

### **International Project to Develop Potato Tuber Moth Resistant Potatoes (USAID)**

Potato tuber moth, *Phthorimaea operculella* (Zeller), is the most serious insect pest of potatoes worldwide. The introduction of the *Bacillus thuringiensis* (Bt) toxin gene via genetic engineering offers host plant resistance for the management of potato tuber moth. The primary insect pest in Egyptian potato production, like many other countries in the Middle East, is the potato tuber moth. In the field, the moths lay their eggs on the potato foliage and the hatched larvae mine the foliage and the stems. This feeding damage leads to irregular transparent tunnels in the leaves and weakening of the stem. The larvae attack the tubers through infected stems or directly from eggs, which are oviposited on exposed tubers or where soil cracks allow moths to reach the tubers. Larvae mine the tuber in the field and in storage reducing potato quality and increasing the potential for pathogen infection. Field and storage studies were conducted to evaluate *Bt-cry5* potato lines for resistance to potato tuber moth in Egypt under natural infestations and their agronomic performance in both Egypt and Michigan. From 1997-2001, field experiments were conducted at the International Potato Center (CIP) Research Station, Kafr El-Zyat, Egypt and/or Agricultural Genetic Engineering Institute (AGERI), Giza, Egypt to evaluate resistance to tuber moth. A total of 27 *Bt*-transgenic potato lines from six different Bt constructs were evaluated over a five-year period. Following harvest and evaluation of the agronomic trials, storage evaluation of potato tuber moth damage was done at the CIP Research Station. The 1997 field trial was the first field test of genetically engineered crops in Egypt. Field tests to assess potato tuber moth resistance in Egypt were able to differentiate between the *Bt*-transgenic lines and the non-transgenic lines/cultivars in 1999, 2000 and 2001. The *Bt-cry5*-Spunta lines (Spunta-G2, Spunta-G3, and Spunta-6a3) were the most resistant lines in field with 99-100% of tubers free of damage. In the 2001 storage study, these lines were also over 90% free of tuber moth damage after 3 mo. NYL235-4.13, which combines glandular trichomes with the *Bt-cry5/gus* fusion construct also, had a high percentage of clean tubers in the field studies. In agronomic field trials in Michigan from 1997-2001 the *Bt*-transgenic lines in most instances performed similar to the non-transgenic line in the agronomic trials, however in Egypt (1998-1999) the yields were less than half of those in Michigan. Expression of the *Bt-cry5* gene in the potato tuber and foliage will provide the seed producer and grower a tool in which to reduce potato tuber moth damage to the tuber crop in the field and storage.

Two transgenic 'Spunta' clones, G2 and G3, produced high control levels of mortality in first instars of potato tuber moth in detached-leaf bioassays (80 - 83% mortality), laboratory tuber tests (100% mortality), and field trials in Egypt (99-100% undamaged tubers). Reduced feeding by Colorado potato beetle first instars was also observed in detached-leaf bioassays (80-90% reduction). Field trials in the U.S. demonstrated that the agronomic performance of the two transgenic lines was comparable to 'Spunta'. We are currently working with USAID, Syngenta and South Africa to commercialize the Spunta-G2 and Spunta-G3 lines.

We have also transformed Atlantic, Lady Rosetta and Jacqueline Lee with the Bt-cry5 gene. We hope to have approval to field test these in Mexico in 2003.

### **Transformation and Evaluation of Potato Cultivars with the *glgC16* (AGPase) Gene**

The processing parameters are strictly defined for potato. For chip processing, a specific gravity of 1.080 is the threshold for processing cultivars. In addition, a low reducing sugar level must occur in the potato tuber at harvest and also during storage prior to processing. Potato breeding of improved cultivars for chip processing has had a low probability of success because of the need to combine numerous economic characteristics into one genotype. In some cases, the genotype may be suitable for chip-processing, but the tuber specific gravity falls below the 1.080 threshold. ADP glucose pyrophosphorylase is an enzyme, which uses the glucose 1-phosphate molecule as a substrate for the biosynthesis of starch. An ADP glucose pyrophosphorylase gene (*glgC-16*) has been isolated from *E. coli* and placed in a plant transformation vector under the control of the patatin promoter. One goal of this study is to examine the value of *glgC-16* to raise the dry matter content for potato tubers.

We have targeted transformation of with the AGPase gene towards lines that have below average solids content. In 2001 and 2002 agronomic field trials were conducted to evaluate agronomic performance, specific gravity, chip-processing, and bruise susceptibility of Onaway, MSE149-5Y and their AGPase transgenic lines. The tuber appearance of the various AGPase lines was similar to non-transgenic Onaway and the MSE149-5Y lines (**Table 4A and 4B**). The results in 2001 and 2002 were, in general, were similar between years. Most of the MSE149-5Y and Onaway AGPase transgenic lines had similar yields, although slightly lower in some lines compared to the non-transgenic parents. In general, the tuber size distribution was quite comparable, although there was a reduction in the number of oversize (>3.25") tubers. The specific gravity for the Onaway and MSE149-5Y AGPase lines was higher than the non-transgenic parents in almost all cases. We also observed a higher incidence of internal defects, specifically hollow-heart, in these AGPase lines. Unfortunately, the results from the blackspot bruise susceptibility tests indicate that the transgenic lines that had higher specific gravities were also had higher blackspot bruise potential (e.g. ONAGP3, ONAGP1, ONAGP2, EAGP24, EAGP4, EAGP9, and EAGP3). We are now making crosses with these AGPase lines to see the effect of the AGPase gene expression on progeny.

## **V. Variety Release**

The MSU breeding program has now named and released its first varieties and is in the process of licensing the new varieties to the Michigan Potato Industry Commission. Three potato varieties were released in 2001: Jacqueline Lee (MSG274-3), Liberator (MSA091-1), and Michigan Purple. MSU is currently licensing these three varieties to MPIC and working out procedures to market these varieties. MSF373-8 is being considered for release in 2003. The named Boulder is being considered because of the large tuber size and low incidence of internal defects.

Boulder is a round white selection with medium specific gravity that can be used in both the tablestock and chip-processing markets. The tubers will chip process out-of-the-field and from 10°C storage. The tubers of Boulder are large in size with a low incidence of internal defects. Boulder was tested in Michigan State University trials, the North Central Regional Trials, on-farm trials in Michigan and other out-of-state replicated agronomic trials. Under irrigated conditions in Michigan the yield is similar to Atlantic, but specific gravity is less. Boulder has a full-season vine maturity that is similar to Snowden, but the tubers size early.

The seedling generation was grown in 1994, followed by two years of selection and seed multiplication at the Lake City Experiment Station, Lake City, MI. Seed increase was located to the Lake City Experiment Station. Since 1998, Boulder has been tested in replicated agronomic trials at the Montcalm Research Farm, Entrican, MI and in the scab nursery at the Michigan State University Soils Farm, East Lansing, MI. In 2000 it was entered into grower on-farm trials in Michigan and the North Central Regional Trial. In 2001 was placed into commercial seed production.

## **VI. Development of a DNA-based Fingerprint System for Potato Varieties**

Since 1990 our potato program has offered a fingerprint service to identify and describe potato varieties. This fingerprint method was based upon isozyme proteins in the potato tubers or leaf tissue. This method has been very reliable, but from a practical point of view, the isozyme protein method requires living tissue to express the proteins. Our goal has been develop a fingerprint system that is DNA-based so that the living tissue requirement would be eliminated. We chose an SSR-based system because of the reproducibility of the PCR-based DNA amplification system. Sixteen potato varieties were chosen for the baseline study. Fifteen SSR primer sets were used. DNA was isolated from fresh leaves, fresh tubers, tuber skins, freeze-dried leaf tissue and freeze-dried tuber tissue. Of the 15 SSR sets, 10 sets amplified readable bands on Metaphor agarose gels that could be used to separate potato varieties. In most cases the varieties could be discriminated with as few as 3 SSR primer sets. Moreover, DNA was able to be isolated from all five tissue sources and obtain repeatable band patterns. This ability to isolate DNA from freeze-dried tissue will allow us to fingerprint varieties when fresh tissue is not available for testing. This SSR fingerprint system can be used alone or in combination with the original isozyme fingerprint system.

**Table 1. Potential Lines for 2003 On-Farm Grower Trials**

Line	Pedigree		Comments
	Female	Male	
<b>Tablestock</b>			
JACQUELINE LEE	Tollocan	Chaleur	Late blight resistant, oval yellow
MICHIGAN PURPLE	W870	Maris Piper	Bright purple skin, white flesh
MSE018-1	Gemchip	W877	Also storage chipper
MSE192-8RUS	A8163-8	Russet Norkotah	Scab resistant russet (Norkotah replacement)
MSE202-3RUS	Frontier Russet	A8469-5	Scab resistant russet
MSE221-1	Superior	MS700-83	Scab resistant (Superior replacement)
MSF373-8	MS702-80	NY88	Chips out of the field, large tubers
MSG050-2	Eramosa	NYL235-4	Flat, round, bright skin
MSH031-5	MSB110-3	MSC108-2	Bright skin
MSI005-20Y	MSA097-1Y	Penta	Yukon appearance
MSI152-A	Mainestay	B0718-3	Late blight resistant, round white
MSJ033-10Y	MSA097-1	Penta	Yellow, Scab resistant
MSJ317-1	B0718-3	Prestile	Late blight resistant, round white
<b>Processing</b>			
MSE018-1	Gemchip	W877	Storage chipper, late
MSF099-3	Snowden	Chaleur	42 °F chipper
MSF373-8	MS702-80	NY88	Chips out of the field, large tubers
MSG227-2	Prestile	MSC127-3	Scab resistant
MSH067-3	MSC127-3	W877	Flat, round
MSH094-8	MSE251-1	W877	45 °F chipper
MSH095-4	MSE266-2	OP	45 °F chipper
MSH112-6	Michigold	Zarevo	42 °F chipper, high solids
MSH228-6	MSC127-3	OP	Scab tolerant
MSH360-1	E55-35	MSF077-8	Scab tolerant
MSI002-3	MSA091-1	MSF134-1	High yield and solids
MSJ080-1	MSC148-A	S440	High yield
MSJ167-1	P84-13-12	MSE250-2	High yield and solids
MSJ453-4Y	Tollocan	MSA091-1	Late blight resistant, yellow
MSJ456-4	Tollocan	Conestoga	Late blight resistant
MSJ461-1	Tollocan	NY88	Late blight resistant

**Table 2. 2001-2002 DEMONSTRATION STORAGE CHIP RESULTS**

Chip Scores represented using SFA Scale†

POTATO LINE BIN#4 [46 °F]	2001		2001 DOH*	2001 SCAB <sup>††</sup> RATING	Sample Dates:							
	DOH*	TOTAL			11/7/2001	12/5/2001	1/1/2002	2/13/2002	3/13/2002	4/10/2002	5/8/2002	6/3/2002
	US#1		SP GR	Bin Temperature (°F)								
MSG227-2	403	449	1.073	0.3	57 °F	56 °F	47 °F	48 °F	47 °F	50 °F	56 °F	62 °F
MSH094-8	370	420	1.073	1.3	1.5	1.0	1.5	1.0	1.0	1.5	2.0	1.5
MSH095-4	444	496	1.080	0.7	1.0	1.0	1.5	1.5	1.5	1.5	2.0	2.5
MSH098-2	344	381	1.074	1.0	1.5	1.0	1.5	1.5	1.5	1.5	2.5	2.0
DAKOTA PEARL	320	407	1.069	0.7	1.5	1.0	1.5	2.5	1.5	2.0	2.5	2.5
<b>SNOWDEN</b>	<b>396</b>	<b>458</b>	<b>1.076</b>	-	<b>1.0</b>	<b>1.0</b>	<b>1.0</b>	<b>1.0</b>	<b>1.0</b>	<b>1.5</b>	<b>2.5</b>	<b>2.5</b>
W1386	345	436	1.073	1.5	1.5	1.5	1.5	2.5	2.0	2.0	1.5	2.5
BIN#5 [52 °F]					57 °F	56 °F	52 °F	52 °F	54 °F	54 °F	55 °F	62 °F
LIBERATOR	395	460	1.075	0.3	1.5	1.0	2.0	2.0	1.5	1.0	1.5	1.5
<b>ATLANTIC</b>	<b>448</b>	<b>491</b>	<b>1.081</b>	<b>1.8</b>	<b>1.5</b>	<b>1.5</b>	<b>1.5</b>	<b>2.5</b>	<b>1.5</b>	<b>1.5</b>	<b>2.0</b>	<b>2.5</b>
MSG015-C	304	384	1.067	1.0	2.0	2.0	2.0	2.0	2.0	3.0	3.5	4.5
MSH067-3	370	420	1.078	2.0	1.0	1.0	1.5	1.5	1.5	1.5	3.5	3.5
MSJ461-1	300	451	1.067	1.0	1.5	1.0	1.5	1.5	1.5	1.0	1.5	1.5
NY120	451	488	1.074	0.3	1.0	1.0	1.0	1.0	1.0	2.0	2.0	2.5
<b>PIKE</b>	<b>355</b>	<b>388</b>	<b>1.080</b>	-	<b>1.0</b>	<b>1.0</b>	<b>1.5</b>	<b>1.5</b>	<b>1.0</b>	<b>1.5</b>	<b>1.5</b>	<b>2.0</b>
LSD <sub>0.05</sub>	63	57	0.003									

†CHIP SCORE: Snack Food Association Scale (Out of the field); Ratings: 1-5; 1: Excellent, 5: Poor.

††SCAB DISEASE RATING: MSU Scab Nursery; 0: No Infection; 1: Low Infection <5%; 3: Intermediate; 5: Highly Susceptible.

\*Agronomic data from Date of Harvest, Round-White Late Harvest (DOH) Trial; Montcalm Research Farm, September 21, 2001.

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



**Table 3. MSU ADVANCED *Bt-cry3A* SELECTIONS PRELIMINARY TRIAL  
Montcalm Research Farm, 2001-2002.**

LINE	2001				2002			
	CWT/A		%		CWT/A		%	
	US#1	TOTAL	US#1	SP GR	US#1	TOTAL	US#1	SP GR
<b>MSE018-1</b>	<b>524</b>	<b>634</b>	<b>83</b>	<b>1.080</b>	<b>377</b>	<b>398</b>	<b>95</b>	<b>1.074</b>
E08.10	491	547	90	1.081	374	400	93	1.078
<b>MSG274-3</b>	<b>287</b>	<b>591</b>	<b>49</b>	<b>1.075</b>	<b>336</b>	<b>432</b>	<b>78</b>	<b>1.071</b>
G38.03	231	530	44	1.073	343	458	75	1.073
<b>ND5873-15</b>	<b>274</b>	<b>311</b>	<b>88</b>	<b>1.077</b>	<b>167</b>	<b>227</b>	<b>74</b>	<b>1.070</b>
ND8.01	306	335	91	1.079	141	178	79	1.067
ND8.04	303	347	87	1.080	178	220	81	1.070
<b>Norwis</b>	<b>379</b>	<b>402</b>	<b>94</b>	<b>1.059</b>	<b>154</b>	<b>165</b>	<b>93</b>	<b>1.054</b>
NO8.03	381	428	89	1.060	177	193	92	1.055
NO8.08	328	354	93	1.063	184	191	96	1.055
NO8.28	372	415	90	1.062	186	194	96	1.056
<b>NY123</b>	<b>424</b>	<b>508</b>	<b>83</b>	<b>1.080</b>	<b>370</b>	<b>400</b>	<b>93</b>	<b>1.074</b>
NY8.10	328	424	77	1.074	313	334	93	1.069
<b>NYL235-4</b>	<b>436</b>	<b>533</b>	<b>82</b>	<b>ND*</b>	<b>312</b>	<b>397</b>	<b>79</b>	<b>1.067</b>
L28.2	294	385	77	ND	288	328	88	1.067
L28.3	316	432	73	ND	301	345	87	1.069
L28.5	308	455	68	ND	292	348	84	1.069
<b>Spunta</b>	<b>292</b>	<b>334</b>	<b>87</b>	<b>ND</b>	<b>313</b>	<b>358</b>	<b>87</b>	<b>1.055</b>
SP8.3	332	423	79	ND	276	315	88	1.056
<b>Yukon Gold</b>	<b>315</b>	<b>350</b>	<b>90</b>	<b>ND</b>	<b>255</b>	<b>282</b>	<b>90</b>	<b>1.064</b>
YG8.8	251	305	82	ND	175	186	94	1.059
YG8.12	265	307	86	ND	151	167	90	1.062
ATLNewLeaf	468	501	93	1.084	321	349	92	1.078
RBNewLeaf	51	258	20	1.075	112	155	72	1.059

Lines are grouped by transgenic parental clone family. Parental clone is bolded.

\*ND: No Data.

**Table 4A. 2001 and 2002 AGPase AGRONOMIC TRIAL, Moncalm Research Farm.**

LINE	2001			2002			2001	2002	2001	2002	2002
	CWT/A		%	CWT/A		%	SP GR	SP GR	HH <sup>1</sup>	HH	CHIP
	US#1	TOTAL	US#1	US#1	TOTAL	US#1					SCORE <sup>2</sup>
<b>ONAWAY</b>											
<b>ONAWAY</b>	<b>423</b>	<b>496</b>	<b>85</b>	<b>324</b>	<b>366</b>	<b>89</b>	<b>1.059</b>	<b>1.057</b>	<b>0</b>	<b>0</b>	<b>3.5</b>
ONAGP2	349	414	84	226	269	84	1.069	1.067	3	7	3.0
ONAGP3	311	373	84	148	183	81	1.071	1.065	1	4	3.5
ONAGP1	301	360	84	283	319	89	1.068	1.067	2	3	4.0
MEAN	346	411		233	272		1.067	1.063			
LSD <sub>0.05</sub>	33	51		57	56		0.002	0.003			
<b>MSE149-5Y</b>											
<b>MSE149-5Y</b>	<b>457</b>	<b>509</b>	<b>90</b>	<b>314</b>	<b>337</b>	<b>93</b>	<b>1.063</b>	<b>1.059</b>	<b>0</b>	<b>4</b>	<b>2.5</b>
EAGP20	431	502	86	281	328	86	1.062	1.062	1	3	4.0
EAGP15	419	481	87	291	335	87	1.063	1.062	0	8	1.5
EAGP4	376	425	89	236	263	90	1.069	1.068	13	22	2.0
EAGP8	360	452	80	308	355	87	1.064	1.062	0	5	2.0
EAGP9	331	369	90	191	215	89	1.070	1.069	29	34	1.5
EAGP24	295	347	85	168	199	84	1.070	1.069	4	16	1.5
MEAN	381	441		256	290		1.066	1.064			
LSD <sub>0.05</sub>	59	53		46	45		0.003	0.003			

Potato lines sorted in decreasing 2001 yield within each parental clone. Parental clone is bolded.

<sup>1</sup>HH: Hollow Heart. Number of tubers out of 40 cut.

<sup>2</sup>CHIP SCORE: Snack Food Association Scale (Out of the field, 9/13/02); Ratings: 1-5; 1: Excellent, 5: Po

2001: Planted May 1, 2001; Harvested September 27, 2001 (150 DAYS)

2002: Planted May 1, 2002; Harvested September 11, 2002 (133 DAYS)

**Table 4B. 2001 and 2002 AGPase SIMULATED BLACKSPOT BRUISE SUSCEPTIBILITY TEST**

LINE	2001							2002								
	NUMBER OF SPOTS PER TUBER						PERCENT (%)	AVERAGE	NUMBER OF SPOTS PER TUBER						PERCENT (%)	AVERAGE
	0	1	2	3	4	5+	BRUISE FREE	SPOTS PER TUBER	0	1	2	3	4	5+	BRUISE FREE	SPOTS PER TUBER
<b>ONAWAY</b>																
<b>ONAWAY</b>	<b>17</b>	<b>7</b>	<b>1</b>				<b>68</b>	<b>0.36</b>	<b>12</b>	<b>9</b>	<b>4</b>				<b>48</b>	<b>0.680</b>
ONAGP2 *	5	8	5	3	3	1	20	1.76	4	5	9	1		6	16	2.240
ONAGP1 *	5	2	9	5	2	2	20	2.12	4	7	4	4	3	3	16	2.160
ONAGP3 *	2	1	4	2	1	15	8	3.76	5	4	3	5	1	7	20	2.560
<b>MSE149-5Y</b>																
EAGP15	16	8	1				64	0.40	20	3	2				80	0.280
EAGP8	15	9	1				60	0.44	12	8	3	1	1		48	0.840
<b>E149-5Y</b>	<b>16</b>	<b>6</b>	<b>2</b>		<b>1</b>		<b>64</b>	<b>0.56</b>	<b>24</b>	<b>1</b>					<b>96</b>	<b>0.040</b>
EAGP20	11	8	4	2			44	0.88	21	1	3				84	0.280
EAGP9 *	4	4	6	3	1	7	16	2.56	2	2	6	8	1	6	8	2.880
EAGP4 *	2	2	6	3	2	10	8	3.24			1	9	4	11	0	4.000
EAGP24 *	4	1	3	3	4	10	16	3.28	5	1	8	5	3	3	20	2.360

Simulated bruise samples were prepared as follows: twenty-five A-size tuber samples were collected at harvest, held at 50 F at least 12 hours, placed in a six-sided plywood drum, and rotated ten times to produce simulated bruising. Samples were abrasive-peeled and scored on October 29, 2001 and October 24, 2002. The table is presented in 2001 ascending order of average number of spots per tuber. Parental clone is bolded.

\*These transgenic lines had higher solids than their non-transgenic parental line.