

Utilization of Genetic Resources



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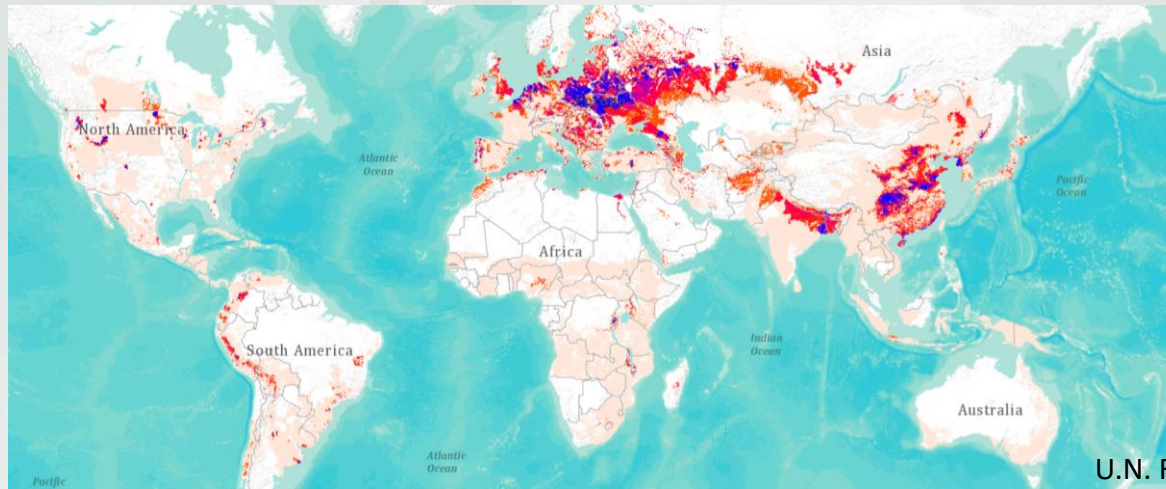
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Potato (*Solanum tuberosum* L.)

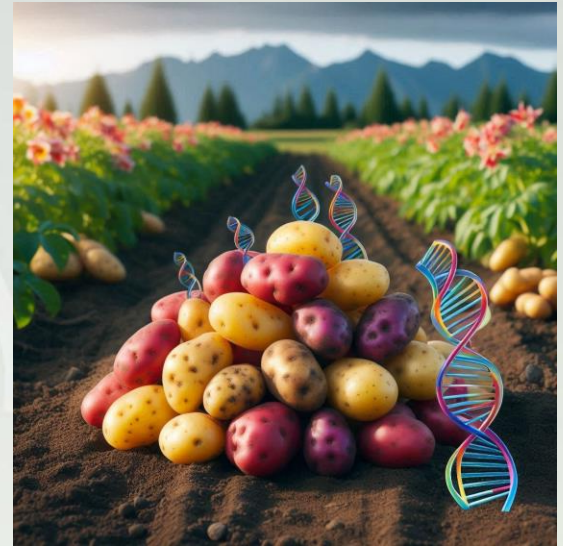
The potato produces more nutritious food, more quickly, on less land, and in harsher climates than any other major crop.

- Fourth in world production
- Critical to local and global food security.
- Potato provides substantially more food per unit of land area compared to other staple crops (Kaldy, 1972; Brown 2005; Zehara 2012).



Potato Genetic Resources Overview

- Diploid potato breeding
 - New potato breeding paradigm
 - Access wild species resources
- Potato biotechnology
 - Traditional genetic modification for certain traits
- USAID Feed the Future Project
 - Late blight resistant (3 R-gene) potatoes



AI image with DALL·E 3

The Potato gene pool

- ~100 tuber-bearing *Solanum* species, 7 cultivated
 - Largely untapped
 - EBN1 species barriers
- Polyploid series (2x - 6x)
 - Majority are diploid ($2n=2x=24$)
 - Cultivated potato tetraploid ($2n=4x=48$)

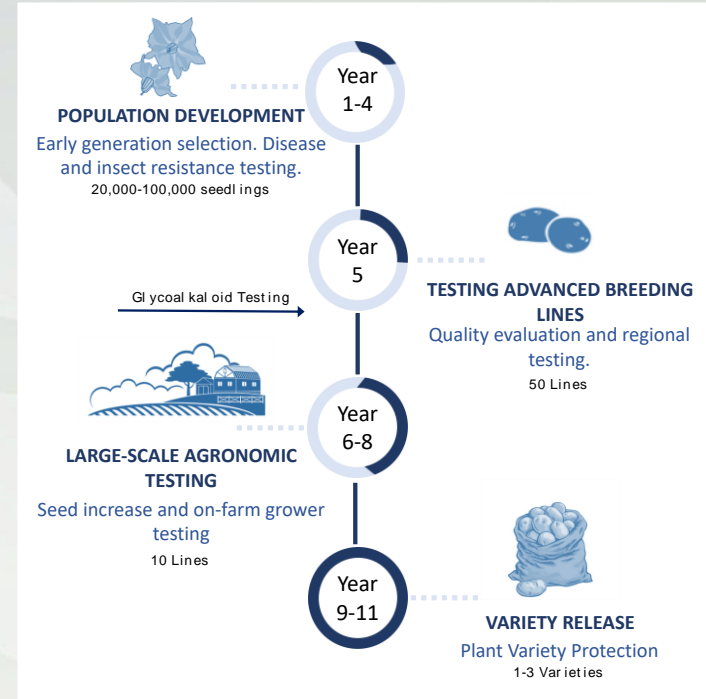


Dr. David Spooner



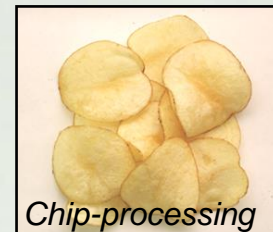
Potato and other vegetative crops have unique challenges to breeding improved varieties

- Long breeding cycle
- Highly heterozygous parents
 - Each cross segregates for all desirable and market limiting traits
 - Polyploidy prevents the fixation of desirable traits in variety development
- ***Difficult to purge undesirable alleles from crop wild relatives***



Priority Potato Breeding Traits

- Disease and Insect resistance (field nurseries)
 - Common Scab (*Streptomyces scabies*)
 - PVY
 - Late Blight (*Phytophthora infestans*)
 - Colorado potato beetle (*Leptinotarsa decemlineata*)
- Storability and chip-processing
 - Storage Sugar Profile
 - 8 months (or more) storage for processing
 - Blackspot bruise
- Adaptability
 - Regional
 - National
 - International



Reinventing diploid potato breeding with self-compatible germplasm

- **Dihaploids** can be extracted from tetraploid germplasm
- **Self-compatible donors** have been identified
- Breeders can access **breeding strategies** not currently feasible for tetraploid potato (F1 hybrids)
 - The breeding process is **more efficient and effective**
- More feasible to **study, access and exploit species** for breeding
- Able to access **genomic tools** more effectively



Dr. Shelley Jansky



Tubers of a diploid interspecific hybrid.

Self-incompatibility, reproductive fitness and genetic load in diploid potato are biological obstacles to inbred line/F1 hybrid breeding

- Most diploids are self-incompatible
- Severe inbreeding depression
 - Limits the number generations of selfing
- Male and female sterility in dihaploids and inbreds
 - Male sterility is almost 100%
 - Female sterility higher than anticipated

Diploid Breeding at Michigan State University

- Crosses with Dihaploids – chip, red, russet, yellow
- Self-compatibility
 - Recurrent selection
 - Backcross introgression
- Selfing – S1, S2
- Breeding crosses
 - Clonal varieties
 - F1 hybrid varieties
- Wide crosses – introgress 1EBN species for resistance traits



MSU diploid (2x) selection

Building the MSU Germplasm base: Dihaploids from cultivated 4x potato

- **Dihaploidization** is a *critical genetic sieve* to reduce the number of deleterious alleles from the tetraploid maternal parent
 - Dihaploids from >50 tetraploid varieties and breeding lines
 - ~1,000 dihaploids generated – 100 female fertile at MSU
 - Potato 2.0 has 100 female fertile dihaploids sequenced
- **Key traits:**
 - Chip-processing
 - Specific gravity
 - Scab resistance
 - PVY resistance
 - (3 R-genes)
 - PLRV resistance
 - PVX resistance
 - Late blight resistance
 - (3 R-genes)
 - Golden nematode resistance



Atlantic-DH120



MSR127-2-DH01

Kate Shaw



Chen Zhang

Developing self-compatible diploid germplasm with a multi-species recurrent selection population

FOUNDERS

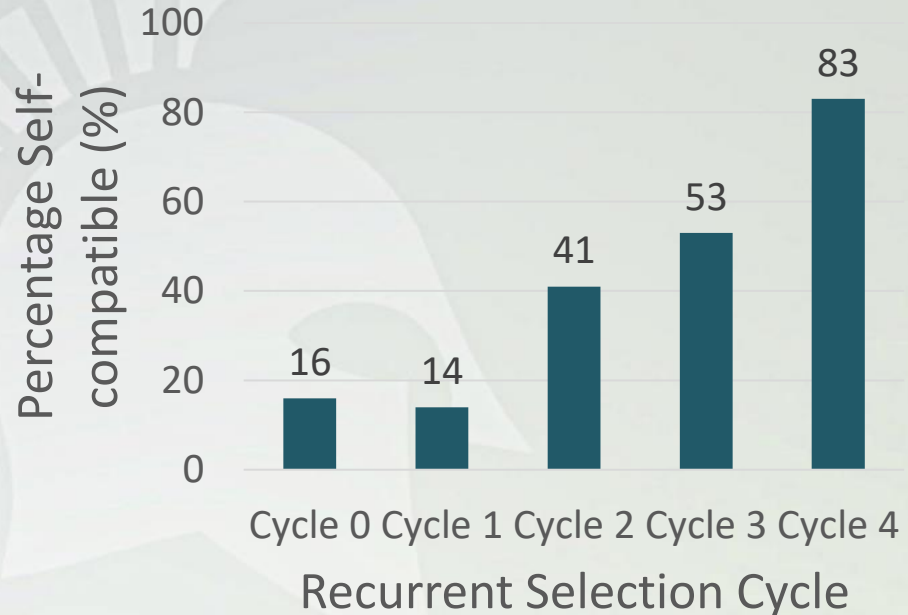
SC Donors

S. chc M6
S. chc 524-8
 XD3
 DMRH-89
 Scab4-48

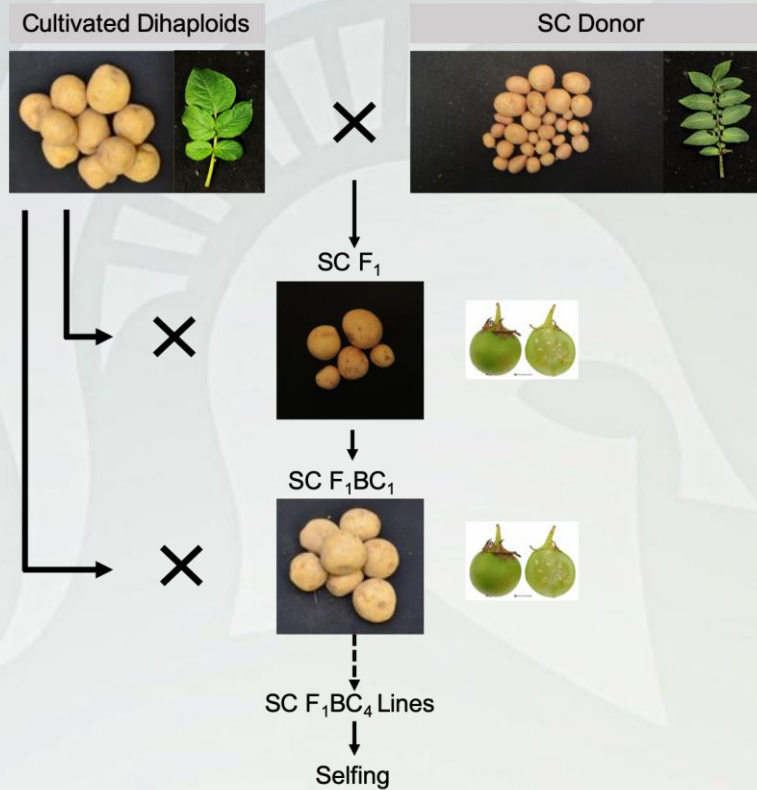
S. chc
S. phu
S. ber
S. tbr
S. mcd



Dr. Maher Alsahlany

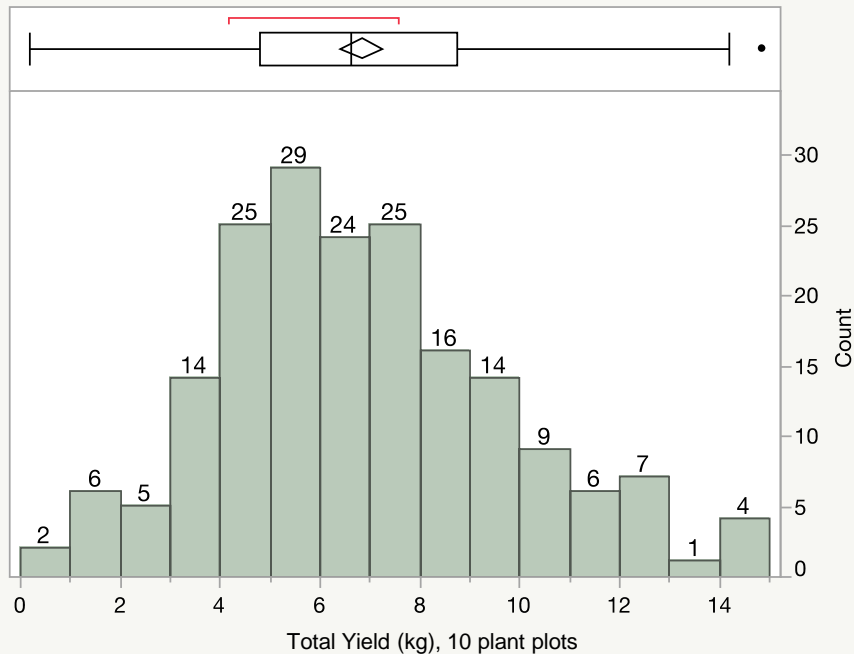


“Backcross” breeding to introgress self-compatibility (SC) into *S. tuberosum* dihaploids



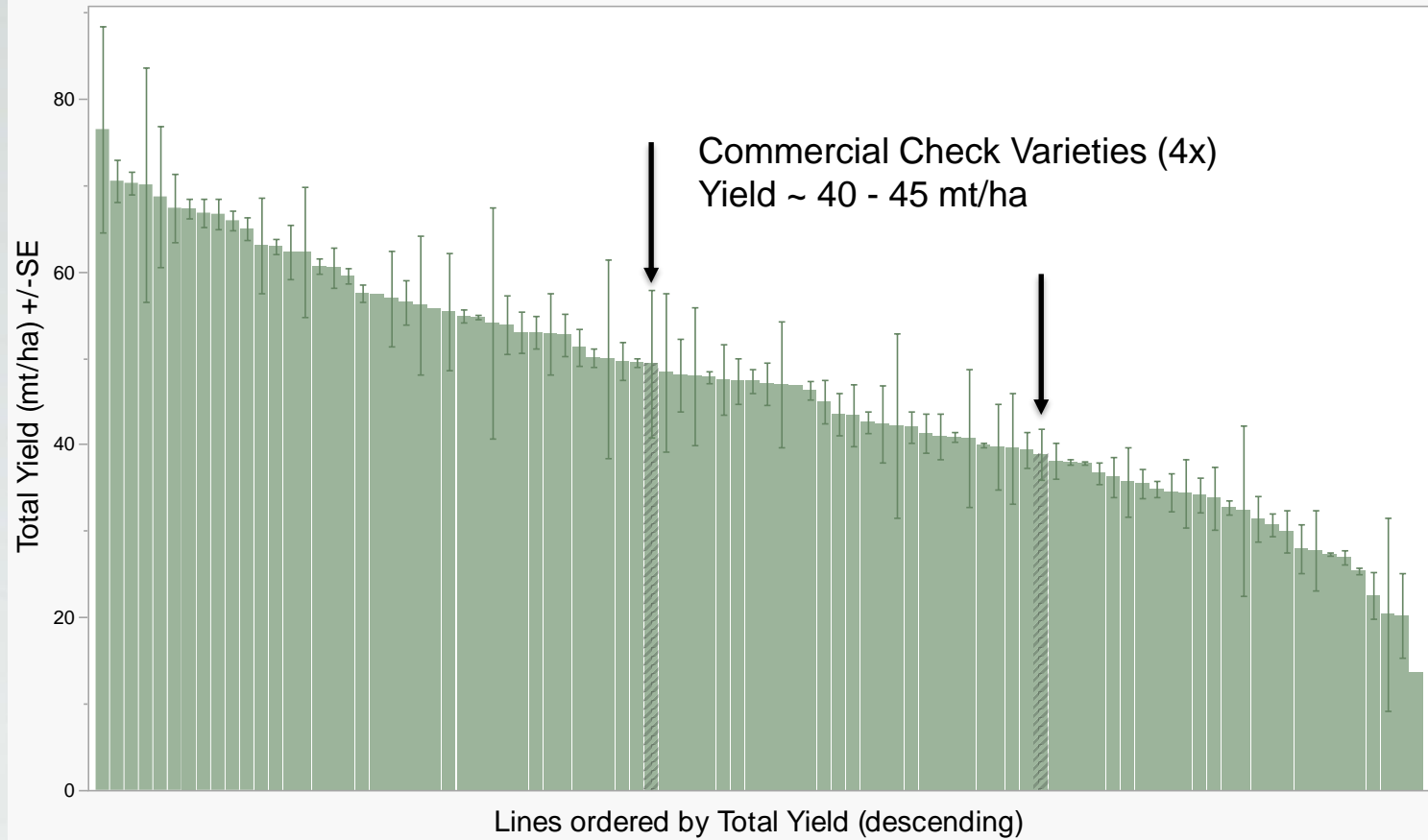
Field Trial of Diploid Clones at MSU

- Total Yield (kg) 2021



Diploid GWAS trial harvest 2021

MSU Diploid Advanced Selections Field Trial

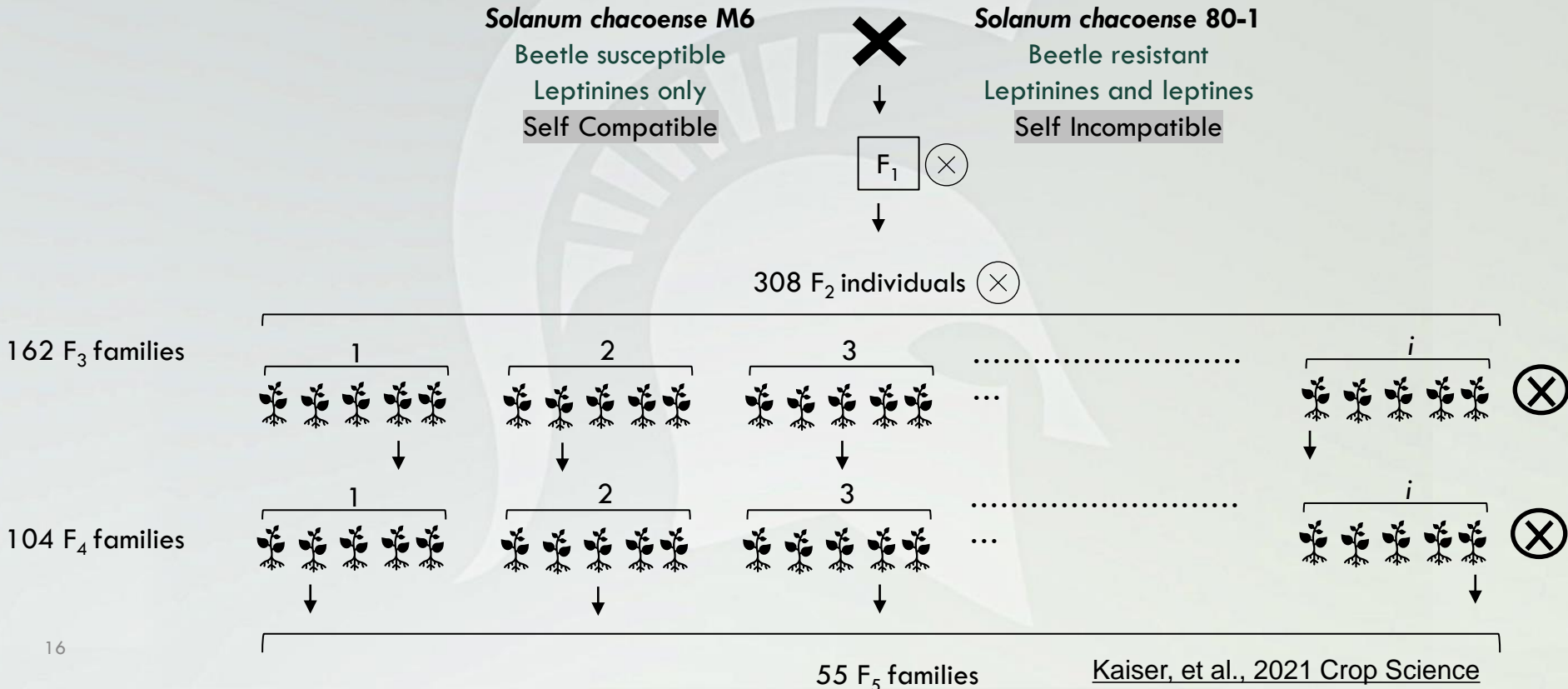


Diploid tuber samples



We are selecting diploids with tuber size and yield comparable to tetraploids.

Creating an intraspecific *S. chacoense* recombinant inbred line (RIL) population for CPB resistance mapping



F4 and F5 inbred lines resistant to the Colorado potato beetle identified in the field



June 23, 2020



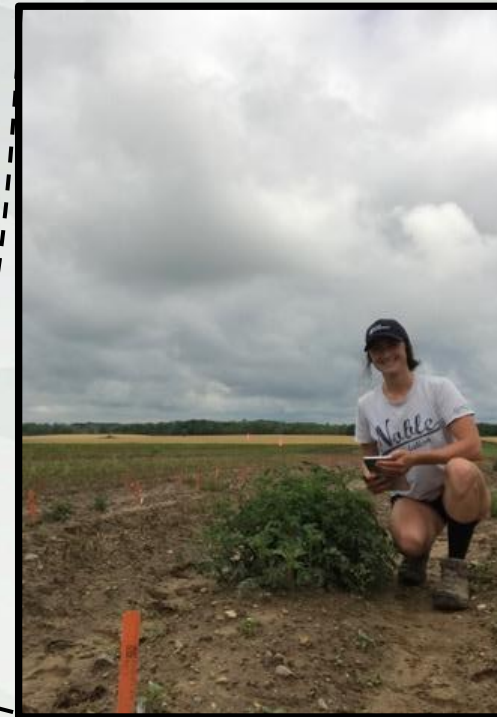
July 3, 2020



July 9, 2020



July 16, 2020



Example of a Colorado potato beetle resistant line

Introducing host plant resistance into cultivated backgrounds

Self-Compatible Colorado Potato Beetle Resistant F₅ Inbred Lines



EE501F5_431

Diploid selections with desirable tuber traits



×

F₁ hybrid

Atlantic (S check)

USDA8380-1 (Resistant check)





Breeding species hybrids
with Colorado potato
beetle resistance

MSII1830-1
((VER x JAM) x MSFF751-01)

Solanum microdontum ($2x=2n=24$)

- 117 accessions in US Genebank
- *S. microdontum* was central in the domestication and improvement of potato (Hardigan et al. 2017)
- Resistance to:
 - *Phytophthora infestans*, (late blight)
 - Potato Virus Y
 - Drought and heat
 - Potato Leaf Roll Virus
 - Bacterial wilt and soft rot
- Six *S. microdontum* genotypes are resistant to four isolates of late blight.
 - Ten genotypes resistant to late blight US-23.



Backcross breeding with *S. microdontum* for late blight resistance

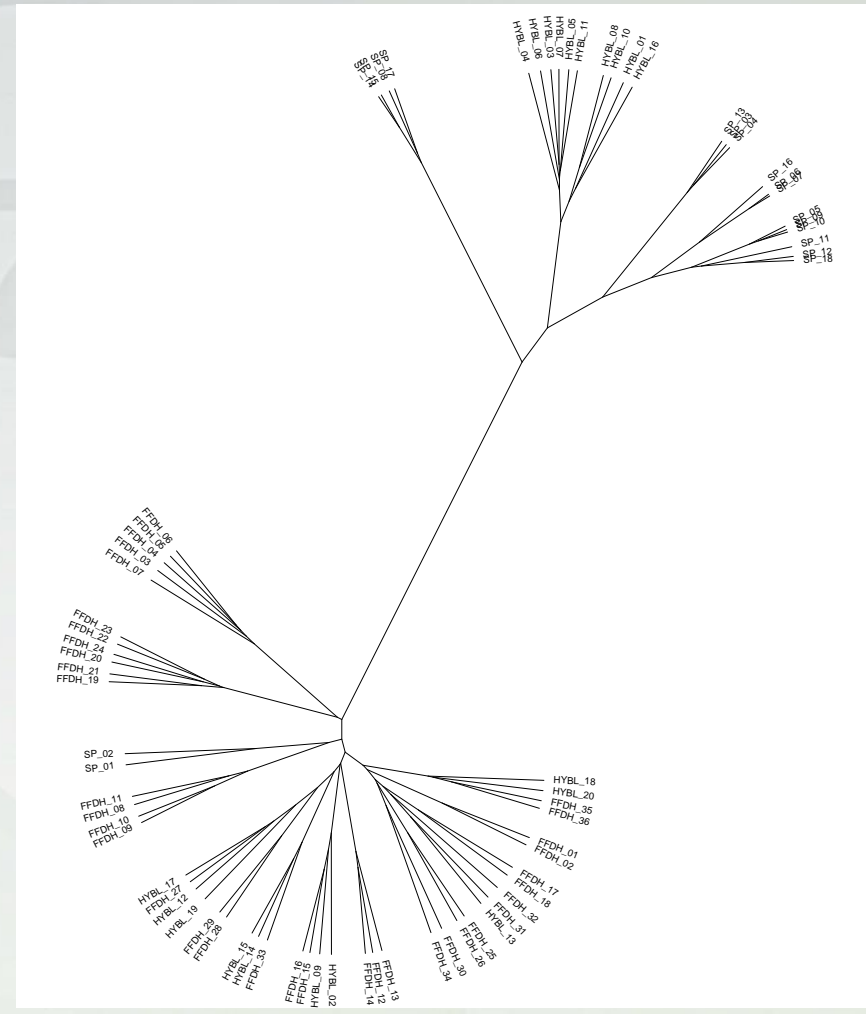
- All BC1 selections were resistant in field conditions to US23
- Most BC1 lines produced tubers
- Some BC1 lines produced fruit in field
- Noted susceptibility to other diseases in some lines
- 2024: BC2 and BC3 crosses for breeding evaluation



S. microdontum BC1

MSU germplasm: Diploids, dihaploids and species

- Female fertile dihaploids
- High yielding MSU Self-Compatible breeding lines from Recurrent Selection and BC breeding
- Introgressed species germplasm
- Potential pools for F1 hybrid breeding

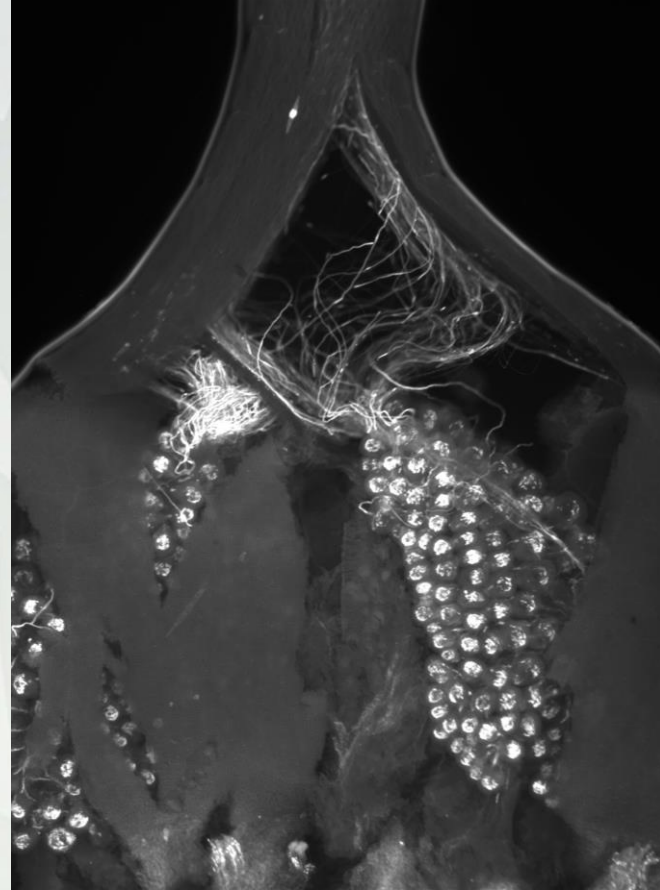
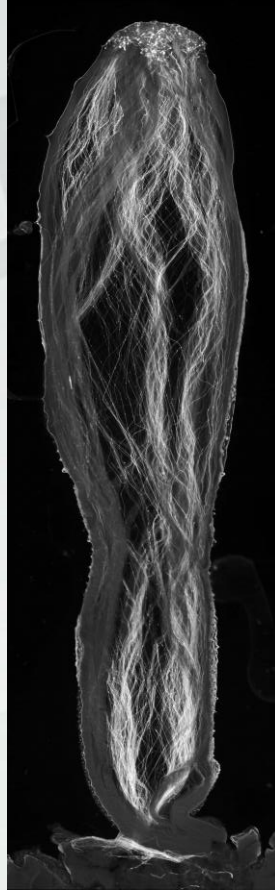


The incredible gains in tomato breeding in the last century are only possible because of its reliable self-compatibility and broad interspecific compatibility

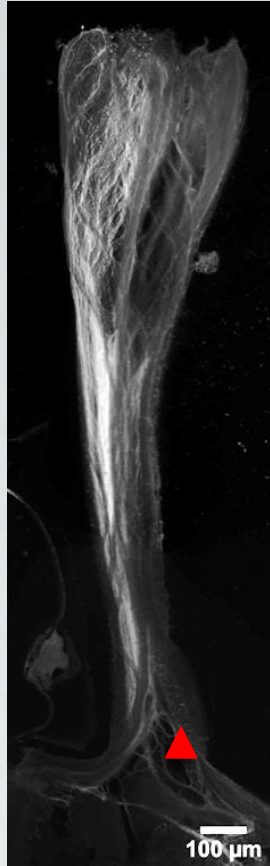
- “Since ca. 1940, breeders have relied increasingly on exotic sources -- particularly related wild spp. -- for desired traits. Since then, accelerated introgression of useful exotic traits contributed to spectacular improvement, manifest in a 4 to 5-fold yield increase. Nearly unknown in tomato cultivars prior to 1940, resistance to at least 42 major diseases has been discovered in exotics and 20 of them bred into horticultural tomatoes -- numbers that are continually increasing.” (Rick and Chetelat, 1995)

Self-Compatibility (SC) in *S. verrucosum*

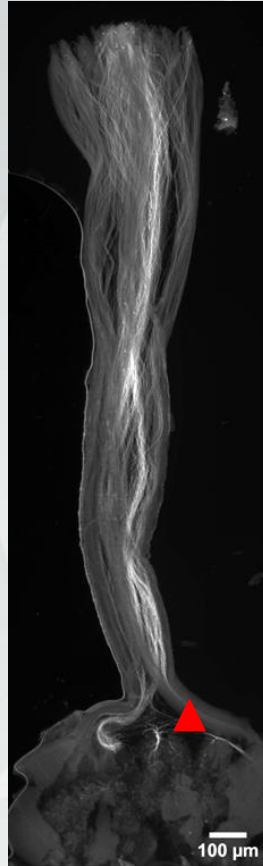
- Why use SvSC?
 - An alternative, complementary SC system to *Sli*.
 - Broad/reliable intra and interspecific compatibility
- Advantages over *Sli*
 - *Sli* can degrade most S-RNase variants, not all.
 - *Sli* does not offer the same level of interspecific compatibility



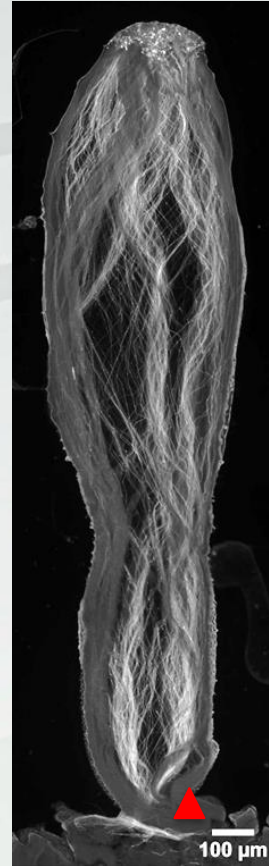
SC in *S. verrucosum* allows for broad interspecific compatibility



S. bulbocastanum



S. commersonii



S. jamesii



S. pinnatisectum

Male vs female side self-compatibility

Female side self-compatibility (FSC) (*S. verrucosum*)

- Robust self-compatibility
- No intraspecific compatibility barriers
- Broad interspecific compatibility
 - Overcome IRBs (like tomato)
- Cytoplasmic male sterility

Male side (*Sli*-based) self-compatibility (MSC) (*S. chacoense*)

- Some robust self-compatibility selections
- Improved intraspecific compatibility
- Limited interspecific compatibility

Will Behling



Mapping Self-compatibility and Interspecific reproductive barriers in *S. verrucosum* - Population Structure

- Parents: *S. tuberosum* x *S. verrucosum*
- F1 hybrid was generated last year: first true F1 hybrid in potato!
- Uniform F1 hybrid progeny



Mapping Self-Compatibility (SC) in *S. verrucosum* – Early findings

- SvSC is a dominant trait.
 - The F1 generation is SC and the F2 generation has a majority of SC individuals.
- SvSC does not require a high degree of male fertility.
 - Pollen stainability amongst SC individuals in F2 ranges from 0.33 to 0.94.
- Too early to confirm this trait is controlled by one locus.
 - Previous literature suggests that SvSC is controlled by a single dominant allele.

Important resistance traits with limited accessibility

- Many “EBN1” species have exceptional resistance to both biotic and abiotic stressors.
- Robust resistance to late blight
 - *S. bulbocastanum*, *S. cardiophyllum*,
S. jamesii, *S. tarnii* and *S. pinnatisectum*
- Resistance to Colorado potato beetle
 - *S. jamesii*, *S. pinnatisectum*



From Nachtinall et al. (2018)



Will Behling

S. verrucosum as a bridge for diploid breeding

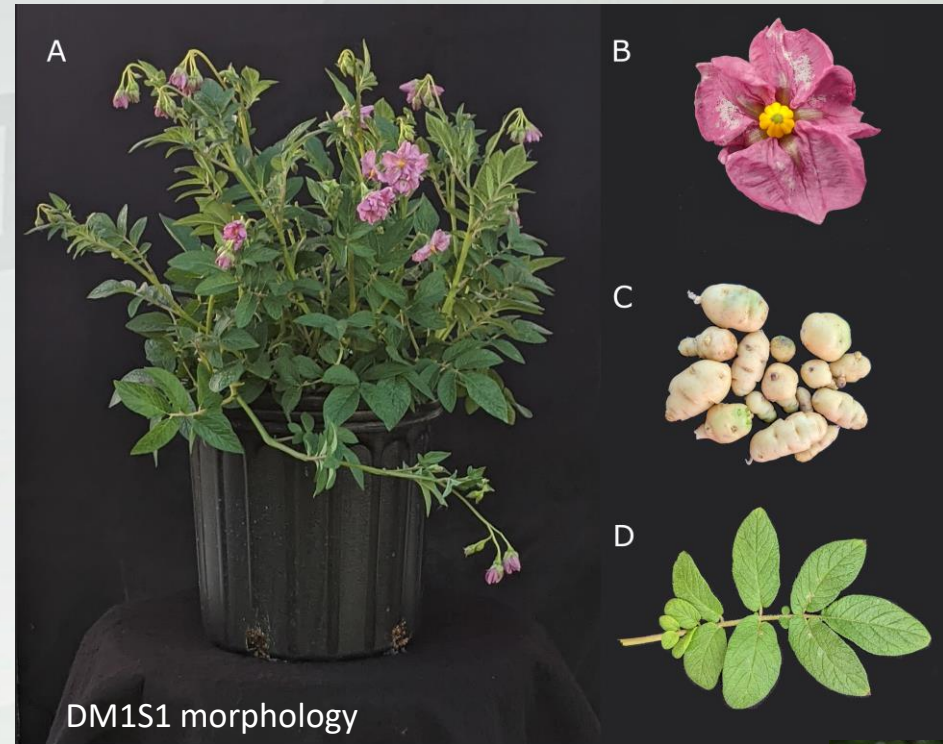
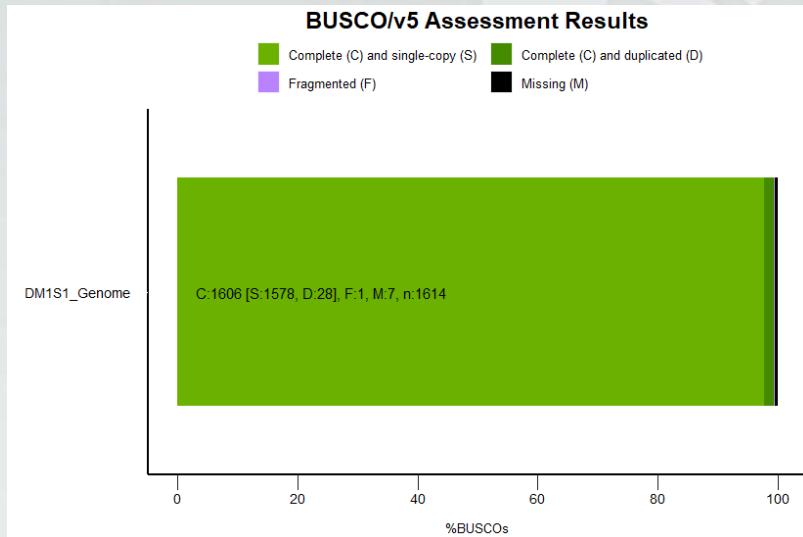
- The use of *S. verrucosum* bridge crosses provides direct access to EBN1 species.
- VER x EBN1 hybrids exhibit low fertility initially.
 - Normal fertility is restored after crossing to cultivated diploids.
- The use of *S. verrucosum* bridge crosses does not require ploidy manipulation or advanced techniques.
 - The resultant diploid hybrids also allow breeding at the diploid level which is more effective in removing undesirable alleles from the species donors via BC breeding.



((VER x CPH) x USW4)

Genomic resources for potato- genome assembly of DM1S1

- DM1S1 is a doubled monoploid derived from 1S1
 - homozygous
 - female fertile
 - produces tubers
 - vigorous
- WGS to resolve haplotype of 1S1

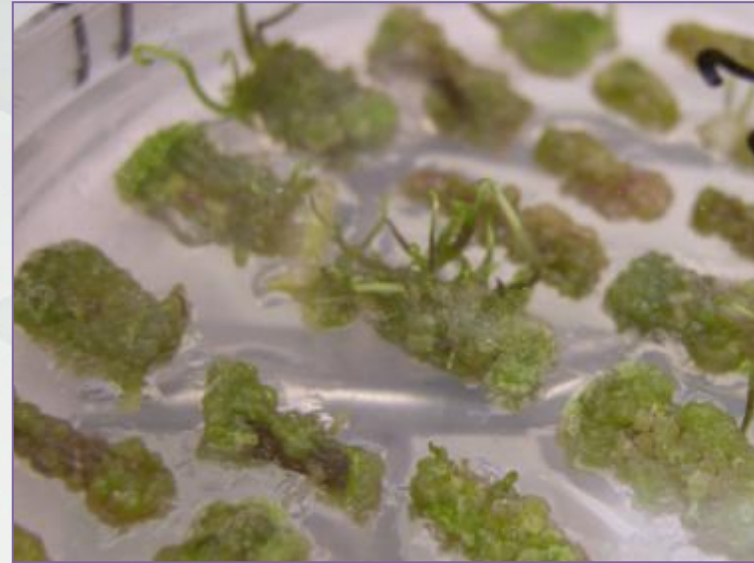


Diploid Potato Summary

- *Potato has an unrivalled wealth of wild species relatives that can transform this crop*
- The diploid (2x) breeding process is more efficient and effective for utilizing genetic resources
 - Self compatibility (*Sli*) introgression
 - Resistance and market quality traits from dihaploids and wild species
- More feasible to study and exploit traits for breeding from wild *Solanum* species
 - We are introgressing insect resistance, LB resistance and virus resistance
 - *S. chacoense*, *S. tuberosum* subsp. Phureja and *S. microdontum*
- Alternate source of self-compatibility has been identified
 - Accessing tertiary gene pool (1EBN) with bridge species (*S. ver*)
 - *S. jamesii*, *S. commersonii*, *S. pinnatisectum*, *S. tarnii*, etc.

Coda: MSU GM Approach: Key Economic Traits of Interest

- Silencing of vacuolar acid invertase (*VInv*) to reduce conversion of sucrose to reducing sugars in potato tubers
- Water use efficiency in potato
- Enhancing Late Blight (*P. infestans*) Resistance of Potato Breeding Lines with three R-genes from *Solanum* species



Silencing of the Vacuolar *Invertase*:

(Can we fix sugar problem in Kalkaska?)

- **Obtained construct** from UW-Madison (Dr. Jiming Jiang)
- **Target Variety:** Kalkaska (MSU potato breeding program)
difficult to transform via *Agrobacterium*
- **Approved for commercial production in 2024 by USDA/APHIS**



Kal.91.03



Kalkaska



Chipped directly after 6 months at 4° C (40° F)

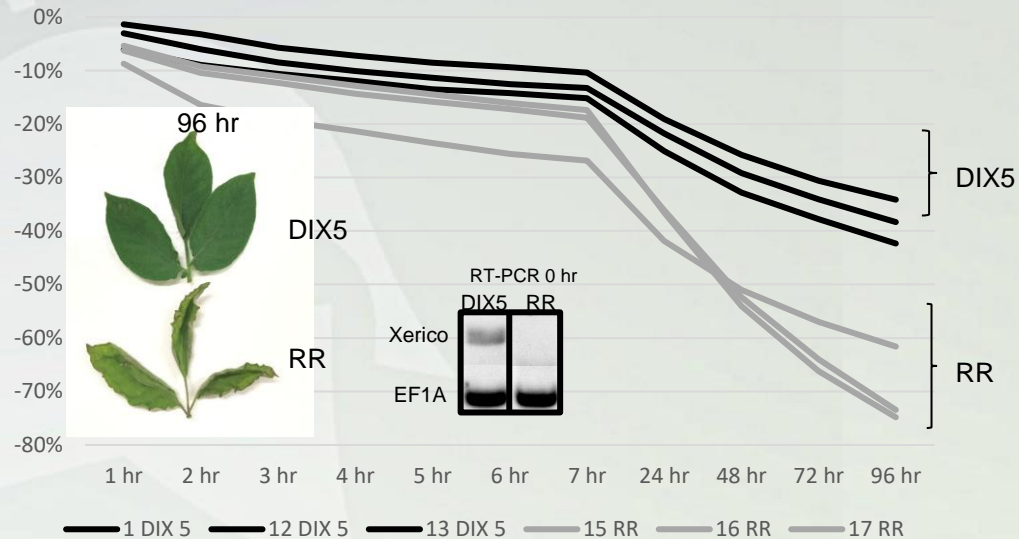
XERICO Drought Tolerance Technology for Potato

A trait for climate change

- Transformations using the XERICO gene coupled with drought inducible promoter in a commercially important potato variety
- Greenhouse studies to verify function
- Field trials to assess agronomic traits

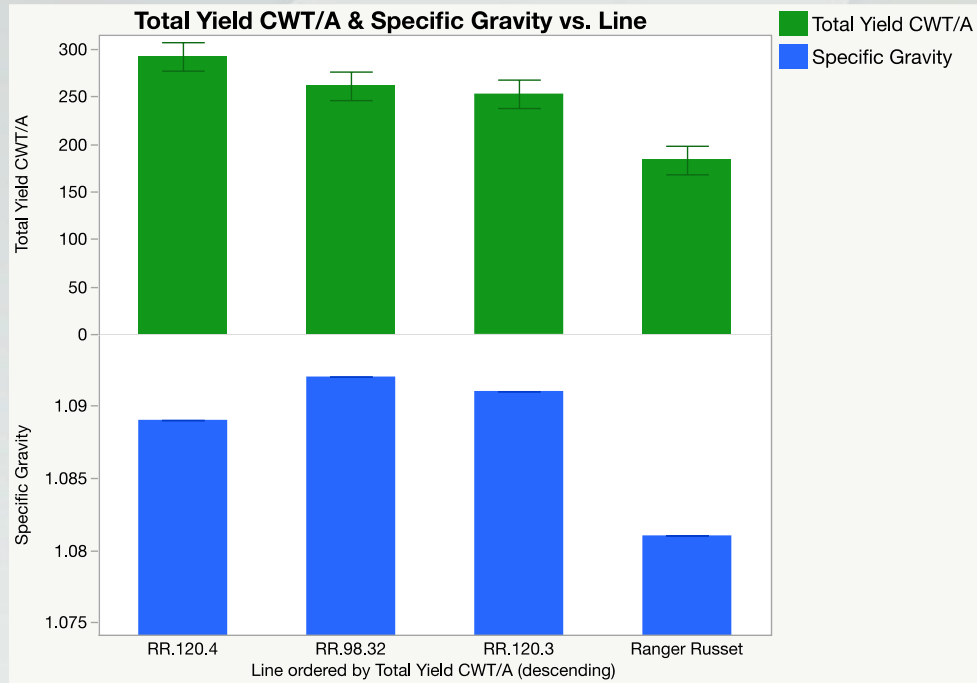
Greenhouse Detached Leaf Drought Test

Percent Water Loss



Drought Trait (XERICO) Field Trials at MSU 2019-23

- Test trait in potato *in situ*
- No yield penalty
- Increase in starch content



The Feed the Future Global Biotech Potato Partnership (GBPP)

- **Partnership:** A five-year multi-institution cooperative agreement between USAID and MSU in partnership with leading international organizations (CIP, AATF), universities, and national research institutions.
- **Goal:** Bring durable biotech late blight resistant potato varieties to smallholder farmers in Bangladesh, Indonesia, Kenya and Nigeria by deploying a 3R-gene strategy in farmer-preferred varieties.



GBPP 3R-Gene Late Blight Resistance Technology

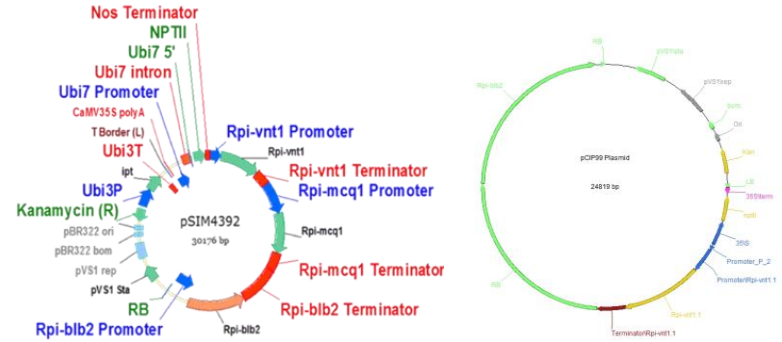
MSU/Simplot Plant Sciences 3R-gene T-DNA (Diamant and Granola)

Rpi-vnt1.1 from *Solanum venturii*
Rpi-mcq1 from *Solanum mochiquense*
Rpi-blb2 from *Solanum bulbocastanum*

CIP-KENYA 3R-gene T-DNA (Shangi and Victoria)

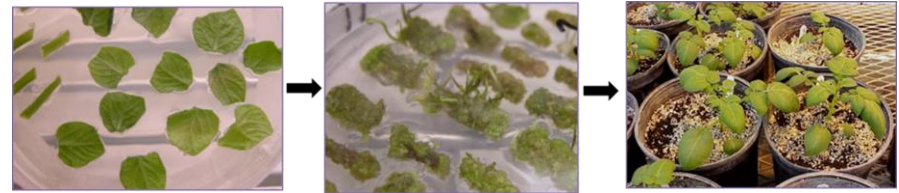
Rpi-vnt1.1 from *Solanum venturii*
Rpi-RB from *Solanum bulbocastanum*
Rpi-blb2 from *Solanum bulbocastanum*

Intellectual Property Rights Acquired



MSU: pSIM4392

CIP: pCIP99



Agrobacterium Potato Transformation

Confined Field Trials for Trait Efficacy and Performance

Indonesia April - July 2021

No Fungicide Treatment



**MSU Granola
3 R-gene
Event
68 DAP**

**MSU
Granola
Control
68 DAP**



**MSU Granola
3 R-gene
Event**

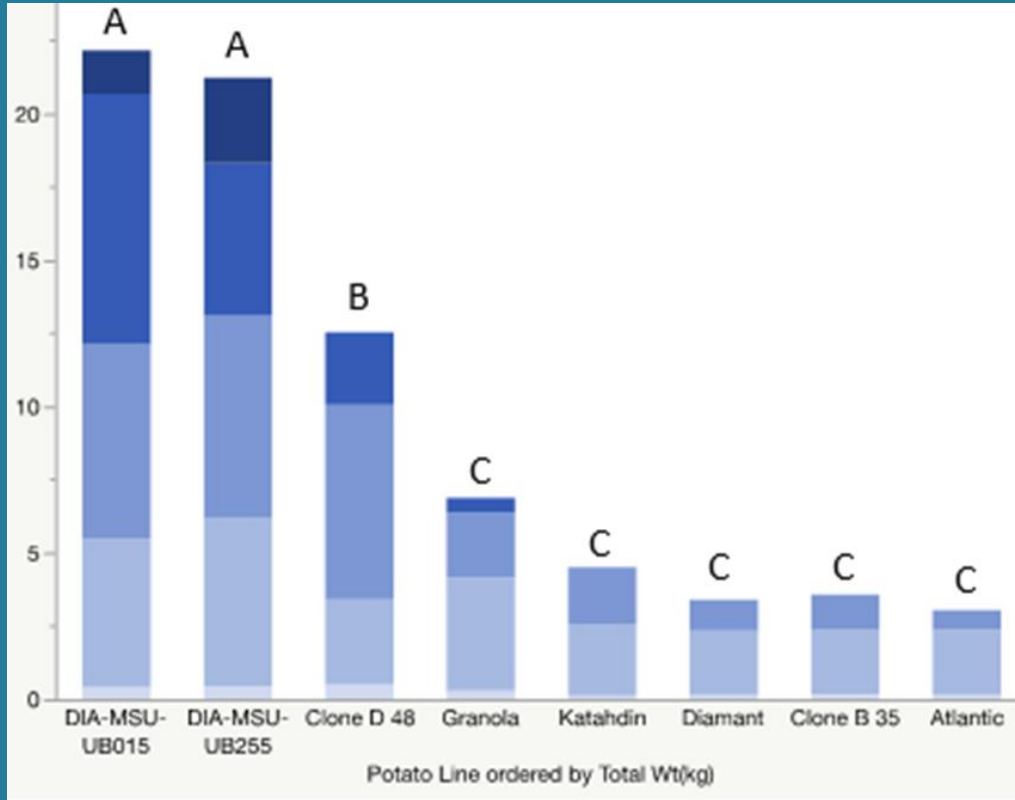
**MSU
Granola
Control**



**MSU
Diamant
3 R-gene
Event**

**MSU
Diamant
Control**

Southeast Asia GBPP LBR Potato Product Yield (kg/plot):



Mean yield and size category of eight potato clones harvested from a foliar late blight trial without protectant sprays. Total mean yields with the same letter are not significantly different as determined by Tukey's HSD ($\alpha = 0.05$).

Acknowledgements



National
Science
Foundation



USAID
FROM THE AMERICAN PEOPLE



Project GREEN



USDA/NIFA
USDA/SCRI



Michigan State University Potato Breeding & Genetics

- <https://www.canr.msu.edu/potatobg>

