Breeding Process Framework: A Powerful Tool for Enhancing Desirable Traits in Potato

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The Potato  
(Solanum tuberosum L.)

- Fourth in world production
- No. 1 Vegetable in the world
- Most popular US vegetable

The potato produces more nutritious food, more quickly, on less land, and in harsher climates than any other major crop.
Potato is the most significant non-grain food crop in the world

http://www.rtb.cgiar.org/RTBMaps/
US Potato Production

- $4 B Annual crop value on 1 M acres
- 5th in world production
- Horticultural crop
  - High cost of production: over $3,000/acre (managing insects, diseases, nutrients and water)
US Potato Production

Data: USDA, NASS 2017

Photo credit: Potatoes USA

- CA: 50,000 + acres
- CO: 25,000 - 50,000 acres
- ID: 500 - 25,000 acres
- ME: 500 - 25,000 acres
- MI: 500 - 25,000 acres
- MN: 500 - 25,000 acres
- MT: 500 - 25,000 acres
Potato consumption in the US

Potato protein: The best plant-based protein

USDA, NASS, 2017
US Potato Breeding is Public

- 9 University breeding programs
- 4 USDA/ARS breeding/research
- 2 Private breeding programs

Public Breeding Programs
Potato Breeding: A challenging 8-12 Year Process

- Tetraploid (4x) and heterozygous
- Prevents the fixation of desirable alleles in variety development
- Each cross segregates for all desirable and market limiting traits

Kaiser et al., 2020 submitted
Potato Breeding Progress has been slow

- For 100 years, potato yield increase mostly due to improvement of production and management techniques.
- Every cross mixes all the important traits for potato agronomic performance and market quality.
Variety change: Top 20 US potato varieties vs maize
Variety adoption is slow

Only 2 of the top 20 varieties were released after 2000.

Russet Burbank is over 100 years old!

Milestones in Potato Breeding and Genetics: Integrating Conventional Breeding, Biotechnology and Genomics

- Conventional Tetraploid (4x) Breeding
  - Vegetatively propagated
  - Since 1850s

- Genetic Engineering
  - 1995

- Diploid Self-Compatibility
  - 2005

- Potato Genome Published
  - Genomic tools
  - 2011

- Diploid (2x) Breeding
  - True seed production
  - 2010

- Gene Editing
  - 2015
Opportunities for Potato in the Genomics Era

- Genome wide markers
  - SNP array technology and other platforms
- Sequencing technologies
  - Assembled genome(s)/annotation
  - RNA-seq
- Genetic engineering
  - Silencing genes
  - New genes for important traits
  - Targeted gene editing
Marker Assisted Breeding – SNP example

- Tetraploid mapping using SNPs to find markers associated with trait
  - Need for 2-4,000 markers to have density for QTL mapping
  - Use of potato browser to develop markers for breeders and annotate genes
- Now have breeder selection tools for many disease resistance and some quality traits

Late blight resistance

Massa et al. G3 | Genes | Genomes | Genetics 2015
Marker Assisted Breeding

- 10 KASP (SNP-based) markers for economically important traits/sample for potato
- High Throughput Genotyping Services (HTPG) is a shared KASP genotyping platform serving CGIAR centers and their partner programs
- HTPG was established to provide low-cost and world class genotyping services to breeding programs working on CGIAR mandate crops, including potato
Genome wide association studies (GWAS)
- understanding contributions to resistance traits in breeding population

Identified SNPs associated with different R-gene hotspots.

Enciso, et al., 2018
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
VInv silencing for improving storability for processing

- RNAi invertase gene silencing events with Kalkaska variety
- Simplot Plant Sciences has released three VInv silenced potato varieties in the US in 2016 that are in the commercial market

Kal.91.03  Kalkaska

Chipped directly after 3 months at 40F
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
2019 XERICO Field Trials at MSU

- Test trait in potato \textit{in situ}
- No yield penalty
- Increase in starch content
Late Blight - Global Severity
Effectiveness of 3 R-gene Late Blight Resistance

- Proof of Concept
- MSU 3 R-gene Events
- Field Trial Test 2017
- Inoculated with US23
- Border rows
  - Atlantic (non-transgenic)
- Center 4 rows include:
  - Single-gene event
  - 3 R-gene events
USAID Feed the Future Biotechnology Potato Project

- **GOAL:**
  - Bring durable biotech late blight resistant potato varieties to smallholder farmers in Bangladesh and Indonesia.

- **VISION:**
  - Project leverages partnerships between private and public sector biotechnology stakeholders
Potatoes feed the hungry
- Bangladesh is 7th in production
- Small holder farmers
- High cost of inputs
- Crop damage by insect pests and disease (particularly late blight)
- Reduction of yield in field and damage in storage
Durable Late Blight Resistance: Stacking multiple genes
3 R-gene potatoes creates a more durable and high level resistance to late blight
3 R-Gene Late Blight Resistant Potatoes for South East Asia

- Indonesian and Bangladesh potato varieties
- MSU received plantlets of 20 selected events of Granola and Diamant from Simplot
- Molecular characterization and confined field trials at MSU
- MSU hydroponic greenhouse minituber production for distributing certified seed
- Field trials to be conducted in 2020 in Bangladesh and Indonesia
“We will be guided by the science-based information, not by the nonscientific whispering of a section of people. As human beings, it is our moral obligation that all people in our country should get food and not go to bed on an empty stomach. Biotechnology can play an important role in this effect.”

- Matia Chowdhury
  Minister of Agriculture
  Bangladesh
Revolutionizing Potato Breeding – *Diploid* Hybrid Potatoes (Potato 2.0)

Comparable with Hybrid Maize Breeding

- **1877**: WJ Beal (Michigan Agricultural College) crosses two open-pollinated maize cultivars to exploit hybrid vigor. He coins the term heterosis.
- **1908**: George H. Shull crosses two inbred maize cultivars to better exploit hybrid vigor.
- **1926**: Henry A. Wallace founds Pioneer Hi-Bred to commercialize hybrid maize.
- **1962**: By 1962, 95% of maize acreage in the US is hybrid maize.
Key Points of Potato 2.0
Diploid Hybrid Potato

- Varieties will be diploid (2x) F1 hybrids
  - Breeders can “fix” weaknesses in a F1 hybrid
- Breeders can access powerful breeding strategies not currently feasible for tetraploid potato
- The breeding process is more efficient and effective
- More feasible to study and exploit traits for breeding
- Able to access genomic tools and gene editing more effectively
Potato 2.0: 2x F1 Hybrid Varieties

- **Advantages:**
  - Simple genetic system for breeding
  - 100 wild species: Sources of economic-related traits
  - Can create inbreds

- **Past Disadvantages:**
  - Gametophytic self-incompatible (SI) system
  - Germplasm unadapted to US

- **Key messages:**
  - Self-compatibility trait from *Solanum chacoense*
  - Breeding will take us from “interesting tubers” to market classes
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MSU Diploid Potato Breeding: Recurrent Selection Breeding Strategy (species-based germplasm pool)

Overall Goal: Develop Diploid Potato Germplasm with Self-compatibility

M6 – self compatibility donor
Improvement of Self-compatibility through Recurrent Selection

Improvement of Self-compatibility through Recurrent Selection

**Self-compatibility Chi-square p=0.0025**
Selections from Five Cycles of Recurrent Selection

Cycle 0
MSBB938-01

Cycle 1
MSCC822-03

Cycle 2
MSDD829-09

Cycle 3
MSEE737-05

Cycle 4
MSFF608-08

Atlantic
4x cultivar
Germplasm Base: Dihaploids from Cultivated Tetraploid Potato (adapted commercial varieties and breeding lines)

Tetraploid (2n=4x=48)

↓

Dihaploid (2n=2x=24)

↓

Male sterile and/or Female sterile

4x Potato Cultivar X Haploid Inducer

U Maine, H. Tan
Dihaploids Extracted from Cultivated Potato at MSU (Past 5 Years)

- **Dihaploidization** is a critical genetic sieve to remove deleterious alleles carried in tetraploid parents
- Dihaploids from 20 tetraploid varieties and breeding lines
- Over 200 dihaploid selections in the breeding pool

- Key traits:
  - Chip-processing
  - Specific gravity
  - Scab resistance
  - PVY resistance
  - PLRV resistance
  - PVX resistance
  - Late blight resistance
  - Golden nematode resistance
Backcross Breeding to Introgress Self-compatibility (SC) into *Solanum tuberosum* Dihaploids (cultivated germplasm pool)
Selections from F1, BC1, and BC2 Generations

**F1**
- MSCC864-17
- MSCC864-19

**BC1**
- MSEE872-03
- MSEE823-05

**BC2**
- MSFF696-01/SC
- MSFF774-04
1980s Version of Diploid Potatoes

International Potato Center
21st Century Version of Diploid Potatoes

Recurrent Selection

Cycle 0 | Cycle 2 | Cycle 4 | S1 | S2

Backcross

BC1 | BC1 | BC2 | BC2
Next Steps: Assessing Heterosis and Inbreeding in the Field for F1 Hybrids

- Crossing between germplasm pools
- 10,000 diploid plants in 2019 field season
- Elite diploid selections with yield and quality
Key Points of Potato 2.0

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Diploid Breeding for Insect Resistance
Breaking through decades of stasis

1980
Host plant resistance to the Colorado potato beetle identified in the wild species S. chacoense

Complex genetics of the trait
Diploid self-incompatibility
Sparse DNA markers

Present
No commercial resistant varieties available
Diploid Breeding for Insect Resistance
Exploiting self compatibility and next generation sequencing

(Kaiser et al., 2020)
Key Points of Potato 2.0

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Gene editing – A powerful tool in potato improvement

- Ability to knock-out and knock-in genes is valuable crop improvement tool
  - Fast changing technology
- Self compatible inbred diploid potatoes may be the best germplasm to exploit gene editing tools for breeding
Self compatibility was achieved in diploid potato lines and edits were transmitted to the progeny.

Self compatible diploid potato fruit

Fruit set in T₀ lines

S-RNase expression after 24 hours post pollination

S-RNase and Cas9 screening in T₁ progeny
Potato Summary

- Potato is most important US vegetable and a world food security crop
- 4x breeding has improved via:
  - Access species germplasm from gene bank to introgress economically important traits
  - Diploid reference genome(s)
  - SNP array technology – marker-assisted breeding
  - Transformation technology – improving market preferred varieties
    - Storage trait, biotic stress, abiotic stress
Potato Summary

- Breeders can access powerful breeding strategies not currently feasible for tetraploid potato
  - Two self-compatible germplasm pools (RS and BC) were generated that are genetically distinct and could be the first step of producing inbred potato lines at the diploid level
  - Two germplasm pools have been selected for agronomic traits
- In the future varieties will be F1 hybrids
  - We have initiated inbred line development and heterosis testing
Potato Summary

- The breeding process is more efficient and effective
  - Breeding cycles for recurrent selection, backcrossing and inbreeding are accelerated (1-3 years vs. 5-8 years)
- Breeders can fix or introduce traits in a F1 hybrid
  - Backcross breeding can introduce traits into inbred line
- More feasible to study and exploit traits for breeding
  - We are introgressing insect resistance via marker-assisted breeding
- Able to access genomic tools and gene editing more effectively
  - Gene editing can be used effectively and efficiently without USDA regulation in many instances
What is next: Potato 2.0 (USDA/SCRI 2019)

- Improve self-compatibility and fertility
  - Combination of breeding and gene editing
- Broaden genetic base
  - Russets, reds, yellows, chippers
- Develop key parental inbreds for market classes
  - Select for tuber traits and maintain fertility
- Research and test seed production systems
Acknowledgements

Table: MSU Potato Breeding and Genetics Program

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