

Exploiting Genetic Variation for Wheat Improvement in the Northern Great Plains

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Research Questions

Crop improvement is predicated on exploiting genetic variation. Without this variation, breeders cannot advance germplasm for any of the important traits of interest to growers. This project seeks to answer the question: what degree can we enhance economically important traits in wheat using diverse germplasm from the USDA Spring Wheat Core Collection.

This germplasm enhancement project is based on nested association mapping (NAM) and was initiated in 2013. It is a long-term and broad-based program that will provide a rich source of genetic diversity for many traits that are or may become important to wheat growers in the region. This includes, but is not limited to: yield, protein content, milling and baking quality, root growth, stand establishment, nitrogen use efficiency, water use efficiency, and disease and insect resistance.

Results

Based on single nucleotide polymorphic (SNP) marker data provided by the Triticeae Coordinated Agricultural Project (TCAP), the Spring Wheat Core Collection held by the USDA-ARS National Small Grains Collection was grouped into four subpopulations based on their degree of relatedness. We then selected 409 accessions that represent the greatest genetic and geographic diversity in the Spring Wheat Core Collection. These 409 accessions were designated as the "Spring Wheat Diversity Collection" (SWDC) and evaluated in the field for various traits.

As expected, a wide range of phenotypic diversity was observed for many of the traits in the SWDC. UM wheat breeder Jim Anderson selected 16 accessions from the St. Paul nursery exhibiting superior phenotypes, i.e. normal heading date, short-stature, good straw strength, disease resistance, etc. Phenotype data collected on the SWDC from the field were collated and analyzed. Twelve of the 16 accessions selected by Jim Anderson, plus 18 additional ones selected based on a) genetic diversity as assayed by SNP markers, b) phenotype data collected from the field, and c) geographic origin comprised the final set of select germplasm for development of NAM populations. These 30 (12 + 18) Nested Association Mapping Parental Selects (NAMPS) were sown in the fall greenhouse for crossing with cultivar RB07, selected by Jim Anderson as the recurrent parent. In December 2013, the first crosses of the NAMPS were made with RB07 in the greenhouse. All but five of these crosses were successful; thus, we will be developing 25 NAM populations from the parents listed

in Table 1. Many of these NAMPS are quite diverse in their morphology compared to RB07 (Figure 1). Crossed seed from these hybridizations were planted in the 2014 fall greenhouse for backcrossing to RB07. This was done to recover more of the superior genetic constitution of RB07 since some of the NAMPS are unadapted to the Midwest production region. About 100 BC₁ crossed seed from each cross was planted in the 2014 fall greenhouse and will be harvested by the end of December. Then, one arbitrarily selected seed (single seed descent) from each about 2,500 BC₁F₁ plants will be grown in the 2014 spring greenhouse and harvested in April 2015.

Preliminary phenotyping data were collected on the NAMPS in the field in 2014 and compared to the recurrent parent RB07. We identified a number of NAMPS that exhibited superior disease resistance (to stem rust; *Fusarium* head blight, and leaf rust) and agronomic (plant height) phenotypes (Table 1). Parents that differ genetically from RB07 for a particular trait can be mapped in the derived NAM population.

Application/Use

The germplasm developed from this project will serve as superior, adapted parental material for regional breeding programs aiming to enhance wheat for many different traits, including but not limited to yield, protein content, milling and baking quality, root growth, stand establishment, nitrogen use efficiency, water use efficiency, and disease and insect resistance.

Material and Methods

To start the project, nearly 2,200 accessions of the Spring Wheat Core Collection were genotyped with 90,000 SNP markers by TCAP and analyzed for their genetic relatedness using Principal Coordinate Analysis (PCA). Then, 409 accessions were selected that represent the greatest genetic and geographic diversity in the Spring Wheat Core Collection. These 409 accessions, designated as the "Spring Wheat Diversity Collection" (SWDC), were evaluated for various traits (i.e. heading date, height, awn length and lodging as well as general disease reactions to stem rust, leaf rust, bacterial leaf streak, and *Fusarium* head blight) in the field at Crookston and St. Paul in 2013. Also included in the nursery as controls were the Minnesota wheat varieties of Linkert, Norden, Rollag, Sabin, and Marshall as well as the selected recurrent parent of

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RB07. In the end, 30 accessions were selected for NAM population development based on: a) genetic diversity as assayed by SNP markers, b) phenotype data collected from the field, and c) geographic origin. NAM population development was started with the first cross of these 30 select lines to the recurrent parent RB07. All but five of these crosses were successful. Thus, the final project will include 25 NAM populations. F_1 's from the first crosses were backcrossed to RB07 to recover more of the superior genetic constitution of recurrent parent since some of the NAMPS are unadapted to the Midwest production region. About 100 BC₁ crossed seeds from each cross were planted in the 2014 fall greenhouse and will be harvested by the end of December. We will continue to advance these populations over several additional generations to achieve greater homozygosity, which is required for future genotyping and phenotyping.

Economic Benefit to a Typical 500 Acre Wheat Enterprise

Cultivars bred with one or more of the enhanced traits derived from the NAM populations will increase profitability for wheat producers in the region. It is important to note that this pre-breeding project has a longer-term horizon for results. In this respect, it is similar to breeding programs since it will take several years before direct economic benefits will be realized by growers.

Related Research (effort)

As the NAM populations are developed, we will evaluate them for many different traits of importance to regional wheat producers, including agronomic traits (heading date, height, yield, lodging, etc.), milling and baking quality (flour yield, protein, absorption, mixing time, loaf volume, etc.), and disease resistance (rusts, root rots, bacterial leaf streak, etc.). Other colleagues in the region also have expressed a strong interest in evaluating the NAM populations for specific traits of importance to their programs. Additionally, to effectively map and transfer genes controlling traits in the NAM populations, genotyping must be done. Currently, genotyping by sequencing (GBS) is the best method for achieving a sufficiently large number of markers for a reasonable cost. I will be seeking other sources of funding to conduct GBS on the NAM populations.

Recommended Future Research

In order to fully realize the great potential of NAM populations for enhancing wheat germplasm in the northern Great Plains region, the full course of population development must be followed. The following scheme would be used to develop the NAM populations before advanced homozygous seed stocks are available for multiple trait evaluation.

Revised Timetable for Nested Association Mapping Population Development of Wheat

2013 Fall GH:

- Plant accessions in August-September, make crosses to RB07 in November and harvest crossed (F_1) seed in December. (*Status: Completed*)

2014 Winter GH:

- Plant crossed (F_1) seed in late December-early January, make backcrosses to RB07 in March, and harvest BC₁ crossed seed in April. (*Status: Completed*)
- Establish a genetically pure seed increase of the original 25 NAM parents. (*Status: Completed*)

2014 Summer field:

- Plant BC₁ crossed seed from each cross combination in the field at St. Paul in April (*Status: Postponed until fall greenhouse season to ensure no populations are lost due to weather-related calamities*).
- Disease assessments of original 25 NAM parents. (*Status: Completed*)

2014 Fall GH:

- Plant BC₁ crossed seed from each cross combination in the greenhouse and harvest in December. (*Status: In progress*)
- Collate and analyze data taken from the field on the NAM parents. (*Status: Completed*)

2015 Winter GH:

- Plant one arbitrarily selected single seed (for single seed descent) from each about 2,500 BC₁ F_1 plants and harvest in April (represents 1st selfed generation).
- Test NAM parents for stem rust reaction at the seedling stage.
- Seed of NAM parents and RB07 will be distributed to cooperators around the region so they can test them for their traits of interest. Parents that differ from RB07 for a particular trait can be mapped in the derived NAM population.

2015 Summer field:

- Plant BC₁ F_2 seed in St. Paul in April, record agronomic trait notes from May to July, and harvest in August (2nd selfed generation).
- Collate all data collected on the NAM parents and RB07 by our cooperators and by us.

2015 Fall GH:

- Plant BC₁ F_3 seed in greenhouse in August-September and harvest in December (3rd selfed generation).
- Test parents for leaf rust reaction at the seedling stage.

2016 Winter GH:

- Plant BC₁ F_4 seed in greenhouse in January and harvest in March (4th selfed generation).

- Distribute seed of populations of interest to cooperators for their field tests.

2016 Summer field:

- Plant NAM populations (and parents) segregating for various traits and obtain year 1 phenotype data from the field.
- Collate all data collected on the NAM populations and parents by our cooperators and by us.

2016 Fall GH:

- Plant BC₁F₅ seed (5th selfed generation), extract DNA from seedlings, and perform genotype by sequencing (GBS) if funding can be procured.
- Plant NAM populations (and parents) segregating for various traits and obtain first experiment phenotype data from the greenhouse.

2016 Winter GH:

- Analyze GBS data for the NAM populations.
- Plant NAM populations (and parents) segregating for

various traits and obtain second experiment phenotype data from the greenhouse.

2017 Summer field:

- Plant NAM populations (and parents) segregating for various traits and obtain year 2 phenotype data from the field.
- Collate all data collected on the NAM populations (and parents) by our cooperators and by us.

2017 Fall:

- Analyze data.
- Identify and distribute advanced lines with enhanced traits to regional breeders for crossing in their programs.
- Write up manuscripts for publication.
- Continue evaluations of derived materials until variety candidates are identified.

Appendix



Figure 1. Diversity of spike types in wheat accessions PI 384403 (top left) and PI 189771 (top right) in comparison with Minnesota wheat cultivar RB07.

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Appendix (continued)**Table 1.** Spring wheat core collection accessions selected as parents for development of nested association mapping populations and selected phenotypic data from 2014 at St. Paul and Crookston.

				STP SR*	STP SR	STP	FHB CRK,	STP
LID	ID1	ID2	Origin	Race MCC	Race QCC	LR	Ave Disease	Plant Height
003	Citr	14819	Eritrea	R	R	R	3.0	93
004	Citr	15006	Nepal	S	S	R	3.5	97
005	PI	62364	Venezuela	R	MR	R	3.5	91
006	PI	153785	Brazil	S	MS	S	3.0	107
008	PI	181458	Finland	S	MS	S	2.0	99
009	PI	189771	Tunisia	S	S	S	4.0	94
010	PI	193938	Brazil	R	R	S	2.5	119
011	PI	199806	Peru	S	S	R	5.0	80
012	PI	205714	Peru	R	R	S	1.0	112
013	PI	213602	Argentina	R	R	S	3.0	110
014	PI	220455	Egypt	R	R	S	2.5	104
015	PI	278392	Palestine	S	S	R	3.0	97
016	PI	282922	Argentina	MR	MR	R	1.5	109
017	PI	344018	Angola	R	R	R	3.0	88
018	PI	345693	Belarus	S	S	S	2.5	115
020	PI	374254	Mali	S	S	S	4.0	81
021	PI	384403	Nigeria	S	S	R	5.0	67
022	PI	430750	Yemen	MR	R	R	5.0	80
023	PI	449298	Spain	R	R	S	4.0	74
024	PI	519465	Zimbabwe	R	R	S	5.0	58
025	PI	519580	Chile	R	R	S	4.0	85
026	PI	520033	Kenya	R	R	R	3.5	91
027	PI	520371	Syria	R	MR	S	4.0	89
029	PI	565238	Bolivia	R	R	S	4.5	82
030	PI	623147	Iran	S	S	S	4.0	110
P1	RB07	RB07	USA	R	R	R	2.5	86

LID=Lab identification number; ID1-ID2=Cereal Investigation number for *Triticum* or Plant Introduction number; Origin=Country of origin; STP SR=Stem rust reaction at St. Paul to races MCCF or QCCJ; LR=Leaf rust reaction at St. Paul; FHB CRK=Fusarium head blight reaction at Crookston; Plant heights at St. Paul are in centimeters.

R=Resistant; MR=Moderately Resistant; MS=Moderately Susceptible and S=Susceptible

FHB rating scale is 1=most resistant and 5=most susceptible