Developing Phenomics and Genomics Tools for Pea Breeding

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Overview

In 2020, dry peas were grown on more than 950,000 acres in the US and had a farm gate value of $212,544,000 (NAAS). With growing interest in plant-based proteins, peas will be increasingly valuable to growers and consumers, and predictive selection methods have the potential to help breeders develop elite cultivars faster and more precisely.

Two prediction models were refined during the summer 2022 field season: marker-assisted selection (MAS) maps the pea genome for quantitative trait loci (QTL) associated with high protein, and genomic selection (GS) seeks to predict the phenotypes of lines from their genotypes.

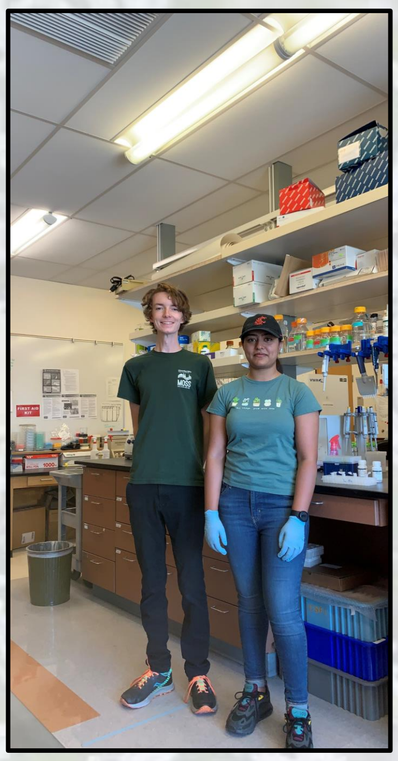
Marker-Assisted Selection

Objectives

- Fine map a genomic region of a recombinant inbred line population for QTL associated with high protein using whole-genome sequencing.
- Develop kompetitive allele-specific PCR (KASP™) markers so that breeders can easily identify high-protein genotypes in the future.

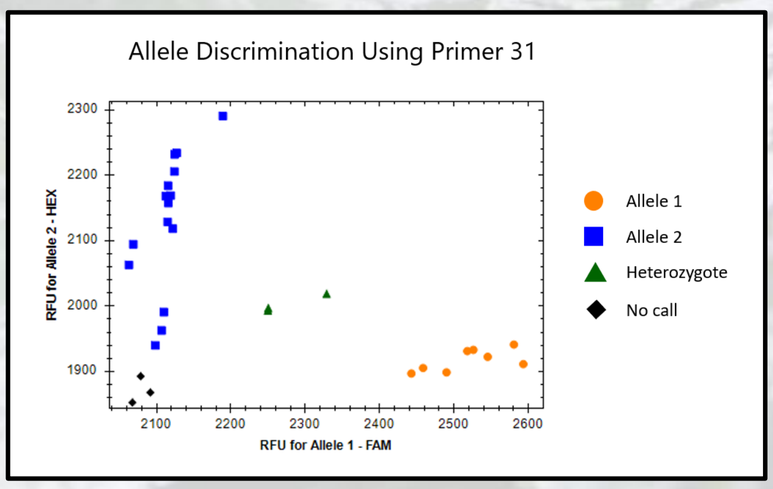
Methods

- An F_6-derived recombinant inbred line (RIL) population was made from the cross Aragorn x Kiflica.
- Protein concentration of seeds was determined with the NIR Matrix-I Spectrometer.
- The 30 highest- and 30 lowest-protein RILs were selected from the population.
- DNA was extracted from young leaf tissue and purified.
- KASP™ markers were used to identify individuals heterozygous within a 2 Mbp segment of chromosome 3.



Results

- RILs 77 and 92 are heterozygous in the 2 Mbp region of chromosome 3. These lines are ideal for fine-mapping the region to identify candidate genes.



Further Research

- Polymorphic single-nucleotide polymorphisms (SNPs) will be used to narrow the 2 Mbp region of chromosome 3 further.
- Candidate genes will be identified using the pea reference genome (Kreplak et al., 2019).

Genomic Selection

Objectives

- Combine a decade of varied phenotypic data with new genotypic data to develop a broad predictive model.

Methods

- Ground truth agronomic measurements have been taken by hand over the 2011-2021 growing seasons.
  - Height
  - Lodging
  - Canopy cover
  - Flowering date
  - Pods per reproductive node
  - Days to maturity
  - Seed yield
  - Seed size
- High-throughput phenotyping data was collected using a UAV during 2021 and 2022 growing seasons.
  - Multispectral images were collected using an AgBot UAV with a mounted 10-band Micasense camera.
  - Images were stitched and analyzed using ImageJ FIJI.

- DNA was extracted and purified for sequencing using genotyping-by-sequencing.
  - 297 breeding lines were phenotyped between 2011 and 2021.
  - 288 breeding lines had viable seeds preserved in the GLGP seed vault.
  - Viable seeds were grown in greenhouse conditions, young leaf tissue was collected, and DNA was extracted.

Further Research

- DNA will be sent to the University of Minnesota Genomics Center for genotyping-by-sequencing.
- After sequence data is available in the fall, genome-wide association analysis will be performed on the data sets using the R and the rrBlup package.

Impact

Both MAS and GS will continue to be refined in the USDA ARS pea breeding program. The plant-based protein fractionation market demands elite pea cultivars with high protein concentration, and tools to create these cultivars, including MAS and GS, will be increasingly valuable in the coming years.

References
Kreplak, J., Madoui, M.A., Cápal, P., Novák, P., Labadie, K., Aubert, G., Bayer, P.E., Gali, K.K., Syme, R.A., Main, D. and Klein, A., 2019. A reference genome for pea provides insight into legume genome evolution. *Nature genetics*, *51*(9), pp.1411-1422.
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