## Brassica carinata crop improvement & molecular tools for improving crop performance

### **Germplasm screening**

- Germplasm collection and diversity
- What has been done in the US to date, plans for future

### Genetic diversity analysis of B. carinata collection

- Genotyping by SSR, genotype by sequencing (GBS)
- Population structure

### Molecular tools for breeding

- Nested Association Mapping (NAM) project underway
- Traits of interest in carinata breeding
- Phenotyping of lines

## B. carinata Germplasm Collection



- Agrisoma collection: > <u>600</u> accessions and breeding lines from eight sources
- AAFC breeding material



## Carinata diversity





## Carinata diversity





### US Carinata germplasm screening

- Most lines that have been yield tested in US have been subset of advanced yield trial lines (origin: AAFC breeding program)
- Agrisoma has/is developing diverse inbred lines that can also be screened in various geographies

### Montana

• Havre, Moccasin, Huntley: 2012 (5 to 25 entries), 2013 (5 entries)

### North Dakota

Minot, Drake 2012 (25 entries); Minot 2013 (5 entries)

### South Dakota

- Bison, Dakota Lakes, Wall 2012 (5 entries); 2013 (5 entries)
- 2014 30 entry trial at 3 sites (AAFC + Agrisoma breeding lines)

### Florida

- Quincy, Winter 2012-13 (6 entries)
- Winter 2013-14 (40 entry trial, AAFC + Agrisoma breeding lines)

### Discussion on germplasm screening?

# Assessing population structure and similarity among lines

• 277 lines analyzed using 76 SSR markers (approx. equal # B and C genome markers)



PCA DICE Genetic diversity analysis



## Genotyping of *B. carinata* collection

- Genotyping by sequencing of close to 500 lines
- Aligned sequence data to diploid A and B genomes (available at time)
- Identified 71,000 raw SNPs
- Selected 6,046 SNP loci
  - minor allele frequency (0.05), less than 30% missing data, read depth  $\leq 4$



Elshire et al, 2011, PloS ONE: e19379; Poland et 2012, PloS ONE: e32253

### Graphical genotypes of B. carinata collection



Milne I., Shaw P., Stephen G., Bayer M., Cardle L., Thomas W.T.B., Flavell A.J. and Marshall D. 2010. Flapjack – graphical genotype visualization. Bioinformatics 26(24), 3133-3134.

# Assessing population structure and similarity among lines



## Practical uses and application





Criteria for choosing parents in cross for DH production: Phenotypic, Agronomic, <u>Genetic</u> data to be used in decisionmaking process

<u>Hybrid development</u>: Initial grouping of accessions and lines for combining ability analysis; will be valuable for identifying patterns of high general and specific combining ability

Decisions on A- and R-line development

**Marker discovery**: Use information to identify and/or develop markers linked to alleles of interest



## Marker Development

- Assessing population structure and relationships among lines
- Currently re-sequencing 20 B. carinata lines for SNP development using the new genome sequence
  - potential Illumina or Affymetrix
    SNP array
- Developed approximately 350 singleplex Kaspar SNP markers mapped across the genome

### BcCtg13840-p369 - mapped to B3



### BcCtg13840-p369 – 96 accessions



## B. carinata NAM population in development

- High allele richness **50 founder lines**
- High mapping resolution **2500 RILS**
- Repeated phenotyping possible
- High statistical power
  - every allele represented many times
- Confounding effect of unlinked genes (false positive) lessened
  - all lines half of background one parent
- Lower impact of genetic heterogeneity
- Crossing to common adapted parent

Marker discovery for *B. carinata* germplasm improvement

### **Phenotyping of NAM lines**

- 2014, 2015 Phenotyping of 50 founder lines + common adapted parent
- 2016, 2017 Phenotyping of NAM population lines locations?

### **Traits of interest**

- Seed oil content
- Earlier maturity
- Yield related components: TKW, seeds per pod, etc.
- Glucosinolate reduction low GSL
- Frost/cold tolerance
- Plant architecture characteristics
- Etc.

### **Discussion on marker discovery**

Interest/opportunities?