Pea heat stress-responsive transcriptome analysis and heat tolerance improvement through marker-assisted backcrossing

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Economic importance of dry pea in Canada

Top five dry pea producing countries

- Canada
- Russian
- China
- France
- India

Data Source: FAOSTAT 2014
Mt: Million tons

Canada pea production distribution in 2016

- Saskatchewan 52%
- Alberta 39%
- Manitoba 9%

Data source: Agriculture and Agri-Food Canada
Warming Canada

Canada mean annual temperature

Canada summer seasonal temperature
Warming Canada


Saskatchewan Pulse Grower
Pea Yield Response to Temperature

Huang, 2016

\[ y = -67.089x + 2166.3 \]

\[ R^2 = 0.6531 \]
Pea Yield Response to Temperature and Precipitation

Heat Stress on Pea

Shortens life cycle  (Huang et al., 2017)

Reduces pollen viability (Jiang et al., 2015)

Induces flower, ovule and pod abortion  (Jiang, 2016)

Lowers individual seed weight  (Huang et al., 2017)
Discovery of Heat Responsive Genes via Transcriptome Profiling

1. *A. thaliana* (Swindell et al., 2007); *Wheat* (Qin et al., 2008); *Barley* (Mangelsen et al., 2011); *Canola* (Yu et al., 2014).


3. The expression of heat responsive genes vary among different species and among different organs within a species.
Two Major Next-generation Sequencing Techniques

Illumina

Roche 454

Voelkerding et al., 2009
Experiment 1: Identification of heat responsive genes in pea via transcriptional analysis
Objectives:

1. Profile pea leaf response to high temperature at the transcription level via Illumina based RNA-seq

2. Compare the heat responsive gene expression between a heat tolerant (CDC Meadow; Warkentin et al., 2007) and a heat sensitive (Nitouche) pea variety

3. Build a heat responsive gene expression atlas in pea
Experiment Protocol

1. mRNA Isolation

2. cDNA Sequencing via Illumina Hiseq 2500

3. Sequence read assembly and gene annotation

Gene expression analysis
Experiment 2: Heat tolerance improvement via marker assisted backcrossing
Plant Materials:

PR-11-2 ♂ X CDC Amarillo ♀
F1

X

F2 X CDC Amarillo ♀
MAS BC1 X CDC Amarillo ♀

MAS BC2 X CDC Amarillo ♀

MAS BC3 X CDC Amarillo ♀

BC4

X

BC4F1
Genome-wide SNP Markers

1. 1536 GoldenGate SNPs were identified (Sindhu et al., 2014).

2. Three hundred and fifty markers were selected based on the polymorphism between cultivated P. sativum including (CDC Sage and CDC Centennial – Parents of PR 11-2) map positions and converted to KASP assays and validated (Warkentin’s lab, unpublished).

3. >50% of the KASP markers were polymorphic between PR11-2 and CDC Amarillo (Kishore et al., unpublished).
Huang et al. 2017
Objectives:

1. Introgress loci conferring long flowering duration from PR-11-2 to CDC Amarillo

2. Identify QTL for heat tolerance between PR-11-2 and CDC Amarillo via the development of the backcrossing population
Field Experiment:

Randomized complete block design with 2 reps

Normal seeding date and Late seeding date

Traits of interest: days to flowering, flowering duration, reproductive node number, pod number, thousand seed weight, yield
Funding: