The Transcriptome of Drought Response in Flax

Raju Datla, Helen Booker, Ioannis Mavraganis, Vivijan Babic, Prakash Venglat
National Research Council Canada Saskatoon
University of Saskatchewan
January 18
PAG 2017, San Diego

Background
- Drought or water limiting condition is frequently encountered stress in crops including flax
- Addressing this challenge requires development of new gene based tools and technologies
- Tolerance to drought stress is complex adaptive response and involves diverse cellular and developmental programs
- To improve drought tolerance in flax will require advanced understanding of these processes

Outline
- Targeted screens to identify genotypes that display drought tolerance in flax;
- Phenotypic, physiological, genomic assessment of promising lines;
- Identify genes and alleles involved in drought tolerance in these lines.

Flax genotypes and cultivars Evaluated
- Bethune, Valour, Sorrel, Andro, Sanctuary, Neela, Plava and Vimy
- Longya and Ningya

Experimental Approach
- Selected genotypes grown in pots with regular watering for 20 days;
- Controlled drought stress by withholding water over a period of 4-10 days;
- The drought response symptoms of leaf wilting and senescence were monitored, visualized and analysed;
- Leaf and root samples collected from treated and control plants for developmental, physiological and transcriptome studies.

Drought Response Phenotypes
Drought treatment response in tolerant genotypes - 4 days

Drought treatment response in tolerant genotypes - 5 days

Drought treatment response in tolerant genotype - 6 days

Drought treatment response in tolerant genotypes – 7 days

Drought treatment response in sensitive genotypes – 4 days

Drought treatment response in sensitive genotypes – 5 days
Drought treatment response in sensitive genotypes – 6 days

The genotypes display differential response to drought treatment (10 days)

Recovery response after drought treatment

- Upon re-watering Santuary, Ningya, and Bethune show complete recovery
- Sorrel, Vimy, and Neela show partial recovery
- Longya show no recovery

Differentially Expressed Genes (DEG) in tolerant and sensitive genotypes
Differentially Expressed Genes in Leaf

16 DE genes with contrasting expression profiles between drought tolerant and susceptible lines.

Differentially Expressed Genes in Root

16 DE genes with contrasting expression profiles between drought tolerant and susceptible lines.

Differential expressed genes in drought tolerant flax genotypes

Differential expressed genes in susceptible flax genotypes

Principal Component Analysis of RNA-seq data collected during the drought and re-watering treatment.

Differentially Expressed Genes (DEG) in tolerant and sensitive genotypes
**Function**

**Description**

- **ABF1** encodes a putative transcription factor that contains a homeodomain closely linked to a leucine zipper motif. Transcript is detected in a variety of tissues and is upregulated in response to cold stress, salt stress, ABA treatment or dehydration.

**Response to various stimuli (including drought)**

**AT5G59320.1** is a member of the Late Embryogenesis Abundant (LEA) proteins which typically accumulate in response to low water availability conditions. It has been shown to promote protection against desiccation in seeds and other plant materials.

**AT1G04110** is a gene encoding a hydrophilic protein lacking glycine-rich domains that is expressed in response to drought stress, light stress and treatment with plant hormones.

**AT1G47960.1** encodes a delta1-pyrroline-5-carboxylate (Pro) synthase that catalyzes the rate-limited enzyme in the biosynthesis of proline. Gene is expressed in reproductive organs and tissues under non-limiting conditions.

**AT1G49720** encodes a member of the bZIP family protein whose expression is induced by ABA. It has been shown to bind to ABA-responsive elements (AREs) and play important roles in carbohydrate metabolism, stress responses and sugar signaling. This protein may inhibit the activity of other proteins.

**AT2G41430.1** encodes a subtilase protein that plays a role in protein turnover in the cytoplasm. It is involved in the degradation of proteins under various stress conditions but in the whole plant under water limiting condition.

**AT2G46680.1** encodes a protein whose expression is induced early on in response to dehydration stress and is involved in the protection against desiccation in seeds and other plant materials.

**AT1G47960.1** encodes a protein whose expression is induced by dehydration and ABA. The mRNA is cell specific and regulated in an ABA responsive manner.

**AT1G54100.1** encodes a protein that is implicated in drought tolerance. This protein may be involved in the regulation of gene expression under drought conditions.

**AT1G01470.1** encodes a protein that is implicated in the response to drought stress. This protein may be involved in the regulation of gene expression under drought conditions.

**AT2G38470.1** encodes a protein that is implicated in the response to drought stress. This protein may be involved in the regulation of gene expression under drought conditions.

**AT1G76180.1** encodes a protein that is implicated in the response to drought stress. This protein may be involved in the regulation of gene expression under drought conditions.

**AT5G50720.1** encodes a protein that is implicated in the response to drought stress. This protein may be involved in the regulation of gene expression under drought conditions.

**AT5G65730.1** encodes a protein that is implicated in the response to drought stress. This protein may be involved in the regulation of gene expression under drought conditions.
Percentage of open stomata in drought tolerant and sensitive flax genotypes

Stomatal response in tolerant genotypes

Stomatal response in sensitive genotypes

Stomatal dynamics in Bethune

Drought treatment (7 days) response associated with stomata in Flax - Sanctuary genotype

Stomatal dynamics in Longya
Soil water potential retained better in tolerant genotypes

Days after treatment

Summary
- Evaluated 10 flax genotypes for drought tolerance
- Performed phenotypic and gene expression studies under controlled greenhouse conditions
- Identified genotypes showing tolerance (e.g., Sanctuary, Bethune, Ningya) and sensitivity (e.g., Sorrel, Vimy, Longya) to drought challenged conditions
- Analysed differentially expressed genes associated with response to drought and identified putative targets
- Stomata associated functions implicated in drought tolerance

Acknowledgements

Bioinformatics: Dustin Cram and Yifang Tan
Collaborators: Mike Deyholos and Sylvie Cloutier
Support: Growing Forward 2 & Flax Council of Canada