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Reducing the Potential for Acrylamide Formation in Wheat Products

Amy Hauver
University of Nebraska-Lincoln, amy.hauver@gmail.com

P. Stephen Baenziger
University of Nebraska-Lincoln, pbaenziger1@unl.edu

Mary J. Guttieri
University of Nebraska-Lincoln, mguttieri2@unl.edu

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Amy Hauver & Dr. P. S. Baenziger & Dr. M. Guttieri

Background

What is Acrylamide?

A *possibly carcinogenic* chemical formed from *free asparagine* during high-temperature cooking of foods containing starch. It is created via the Maillard reaction.

Maillard Reaction

Amino Acid

✚

Reducing Sugar

300 °F

Browning and Caramelization

How can you reduce its formation?

By reducing the level of free asparagine, you are able to correspondingly reduce the potential for acrylamide formation in baked wheat products.

Asparagine production:

Asparagine is an amino acid that plays a key role in nitrogen assimilation, distribution, and remobilization in plants. In developing seeds, it is an important form of amino acid that translocates from the parent plant. There are three metabolic pathways for asparagine synthesis, and if one is shut down, there may be a reduction in overall asparagine production.

Objectives of this study

Short-Term

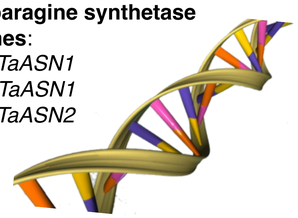
Generate genetic stocks and DNA markers to evaluate the use of knockout mutants in asparagine synthesis pathway.

Long-Term

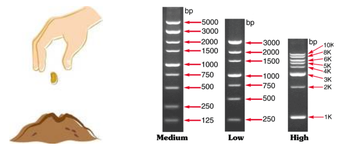
Reduce the level of free asparagine produced in wheat grain.

Methods

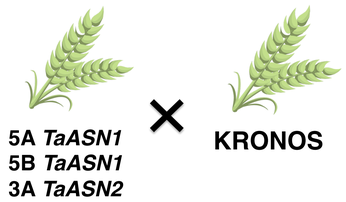
First**, parental seed lines** were acquired with mutations identified in the **glutamine-dependent asparagine synthetase genes**:
5A *TaASN1*
5B *TaASN1*
3A *TaASN2*



Next, **the plants were grown in the greenhouse** and, using the **KASP** genotyping system, **DNA markers were developed** based on the DNA sequence information for these mutations.



Mutants then backcrossed to non-mutagenized (wild type) **KRONOS** (Select progeny **(F1)** with the mutation of interest)



Extract F1 DNA and test it with previously developed markers to ensure crosses were successful
Repeat backcross of F1 plants to KRONOS to develop near-isogenic lines.

Identify F2 seedlings that are **homozygous mutant** and **wild type**. Successful crosses are selected and the plants grown to maturity under controlled fertility condition to ensure **isogenic seed**.

Harvest F2 seed and measure the asparagine concentration in the grain by GC analysis in the Phenomenex EZFast assay to judge success.

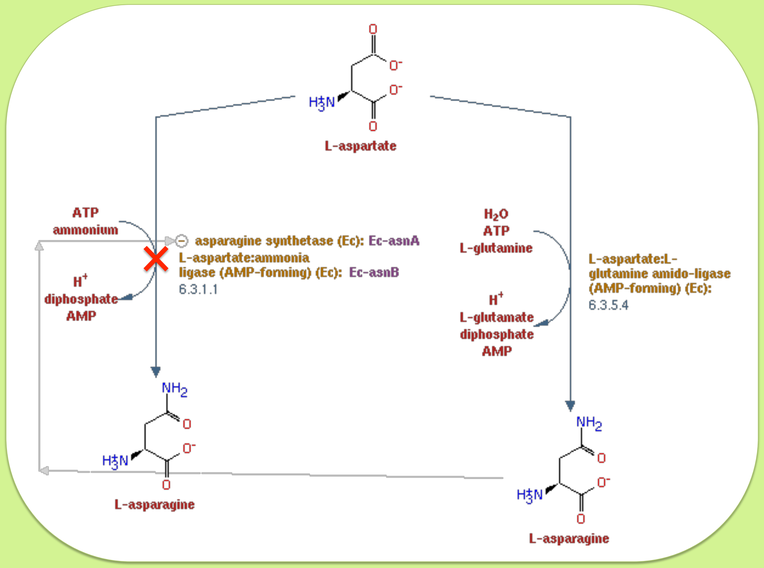


Figure 1: **MetaCyc Pathway**: superpathway of L-asparagine: two of three possible pathways in asparagine production

Anticipated Results

Set-Backs:

Cool temperatures and insufficient lighting in the greenhouse resulted in sterilization of the F1 plants, and we were forced to backtrack and replant.

Revised Plan:

Reapply for Summer 2016 UCARE ✓
January 20, 2016 – Replant ✓
March 25, 2016 – Create F1 crosses ✓
April 25, 2016 – Collect F1 seed and plant
June 25, 2016 – Create F2 seed
July 25, 2016 – Collect F2 seed and analyze

By targeting asparagine synthesis pathway mutations, the data will theoretically show:

1. The shutting down of one pathway in the production of asparagine and a slight decrease in its production, garnering the expected results.
2. The shutting down of one pathway and a corresponding increase in production of asparagine from the others, therefore not gaining the expected results.
3. The shutting down of two or more pathways, adversely affecting the plants growth and yield.

Further Reading:
Halford, N. G., T. Y. Curtis, N. Muttucumaru, J. Postles, J. S. Elmore, and D. S. Mottram. "The Acrylamide Problem: A Plant and Agronomic Science Issue." Journal of Experimental Botany 63.8 (2012): 2841-851.

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