Influence of Genotype and Environment on Isoflavone Content in Adapted by Non-adapted Soybean Crosses in Ontario

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Background

- **Soybeans & Ontario Agriculture**
  - Soybeans are a significant part of Ontario agriculture. Over 2.3 million acres of soybeans were planted in 2005, making soybeans the largest row crop in Ontario (OMAFRA, 2006).
  - Identity Preserved food grade soybeans are part of Ontario's agricultural success story. IP soybeans allow growers the opportunity for increased revenues on their distinct crop.
  - Specifically new traits in food grade soybeans are being developed to increase the utilization of soybean as a food crop, including soybeans with increased isoflavone content.

- **Soybean Isoflavones**
  - Isoflavones are naturally occurring phytoestrogen molecules found in soybeans. Consumption of soyfoods containing isoflavones may benefit human health by reducing the risk of heart disease, specific cancers, and osteoporosis (Setchell, 1998).
  - The demonstrated health effects of soyfoods have created tremendous public interest and increased the demand for soy products.

- **Genotype by Environment Interaction (GEI)**
  - GEI is an important consideration in plant breeding, referring to the differences in response of a genotype to diverse environments (Yan and Kang, 2003).
  - Stability of a genotype, or predictable performance of a genotype across environments, is desired by plant breeders for the trait of interest.
  - Identifying soybean lines with stable isoflavone content across environments is necessary for the development of varieties which can be grown across Ontario with elevated isoflavone profiles.

- **Isoflavone and Protein Content**
  - Isoflavone content may have a significant negative impact on protein content. Preliminary research has suggested that 'high' isoflavone classes of soybean had reduced protein content versus the 'low' isoflavone class (Primomo, 2005).
  - The factors that might effect such a phenomenon are not yet known.

Research Objectives

1. Investigate the influence of the genotype by environment interaction on the stability of seed isoflavone levels of soybean lines from diverse genetic backgrounds grown across different environments.
2. Examine the relationship between seed isoflavone and seed protein content to determine if high isoflavone levels are correlated with protein levels or if the two traits are phenotypically independent.

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Methods

- **Plant Material:**
- The research is making use of a diverse collection of soybean germplasm held by Ridgetown Campus; using lines generated from crosses made in 2000 between RCAT Angora, a high isoflavone variety, and several high yielding soybean varieties.
- *Experiment 1- 'Adapted by Adapted' and 'Adapted by Non Adapted' Cross Study*
  - Includes lines derived from crosses between RCAT Angora and CK-01, varieties both grown in Ontario, and RCAT Angora and the Chinese variety Heinong 35.
  - Lines derived from these crosses, grown in 2004, were analyzed for total isoflavone content and classified as high, intermediate or low. Five lines from each classification were selected for inclusion in the 2005 and 2006 field trials.
- *Experiment 2- Genetically Diverse Study*
  - Lines derived from 10 crosses are included in this study (Table 1).
  - 2004 material was again analyzed for total isoflavone content. Lines were classified as high or low isoflavone. The lines from each cross with the highest and lowest total isoflavone content were selected for field evaluation in 2005 and 2006.

Table 1: Genetically Diverse Experiment- Cross List

RCAT Angora[1] x OAC Arthur[2]
RCAT Angora x PRO 28-53[2]
RCAT Angora x Ivory[2]
RCAT Angora x CK-01[2]
RCAT Angora x S20-F8[2]
RCAT Angora x Tsurkogane[3]
RCAT Angora x DongNong 42[4]
RCAT Angora x Heinong 35[4]
Heinong 35 x Ivory
RCAT Appin x Suzumaru[3]

Variety Notes: [1]high isoflavone Ontario variety, [2]high yielding commercial Ontario variety, [3]Japanese variety, [4]Chinese variety

- **Plot Locations**
  - Both experiments were grown in four locations across Southern Ontario in 2005. In 2006, the experiments were repeated in the same four Southern Ontario locations with the addition of a location in Eastern Ontario and in Quebec.
  - The locations represent a range of growing conditions in terms of accumulated crop heat units (chu) and soil type.
- **Seed Component Analysis:**
  - Oil, protein, total seed isoflavone content and individual isoflavone content is measured using Near Infrared Analysis (NIR).
  - NIR is quick and cost effective, making it possible to analyze the large number of samples in a breeding program.

Results and Discussion

- 2005 saw above average temperatures throughout the growing season at all locations included in the experiment.

Isoflavone and Protein Interaction

- Figure 1 describes the relationship between total seed protein and total isoflavone levels in two separate studies averaged over 4 locations in 2005. The data suggests a moderate inverse relationship between these two seed traits.

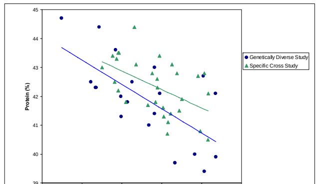

Figure 1: Relationship between total seed protein and total isoflavone in two separate experiments both grown in 2005. Data points are the mean of the values across four locations (RCAT, Woodslee, Inwood, and Exeter)

Results and Discussion

Stability of Isoflavone Content Across Environments

- Within the first year of the study, lines classified as high isoflavone were relatively stable across the wide range of environments, providing evidence that it should be possible to identify and retain high isoflavone lines in a breeding program and in commercial production. (Figures 2a and 2b)

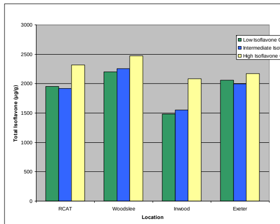

Figure 2a: Mean total isoflavone of each classification at each location in 2005 for the cross RCAT Angora x CK-01.

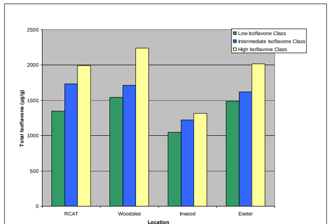

Figure 2b: Mean total isoflavone of each classification at each location in 2005 for the cross RCAT Angora x Heinong35.

- In the genetically diverse experiment, the same isoflavone stability was observed, however there are genetic differences between families. This illustrates that selection of the parents in the breeding program does have significance on the outcome. (Figure 3)

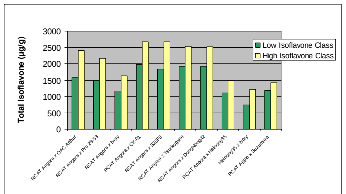

Figure 3: Mean total isoflavone of high and low isoflavone classification lines of each cross grown at all locations in 2005 (RCAT, Woodslee, Inwood, Exeter) .

- A mean/CV stability analysis concurrently assesses mean performance and variability. Lines located within Quadrant I (high mean and low CV) are the desired type. These lines show stability in that they have higher than average performance and lower than average variability over environments. Figures 4a and 4b show lines in both experiments with stable high total isoflavone content, indicating that achieving that goal in a breeding program is possible.

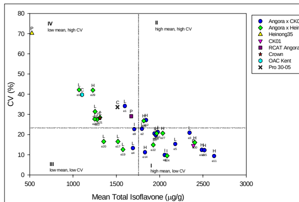

Figure 4b. Mean total isoflavone versus coefficient of variation of 36 soybean lines in an 'adapted by adapted' and 'adapted by non-adapted' cross study conducted across four locations in Ontario in 2005. Entries classified as low are denoted as 'L' and entries classified as high are denoted by 'H'.

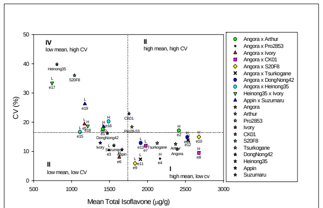

Figure 4a. Mean total isoflavone content versus coefficient of variation of 31 soybean lines in a diverse cross study conducted across four locations in Ontario in 2005. Lines classified as high are denoted as 'H' and lines classified as low are denoted as 'L'.

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