# Relationships of Isoflavone, Oil, and Protein in Seed with Yield of Soybean

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### INTRODUCTION

Soybean is traditionally produced for oil and protein in the seed, which are the economically important seed quality components of the crop. Seed of current U.S. soybean cultivars contains approximately 41% protein and 21% oil on average on a dry weight basis (Hartwig and Kilen, 1991). Smith (1991) summarized American Soybean Association recommendations for the ideal soybean to include high protein, high oil, and very high yield. Because of the positive potential role of isoflavones in prevention of cancer, heart disease, osteoporosis, and menopausal symptoms (Caragay, 1992; Hasler, 1998), some soybean production area may be devoted to contract production for isoflavone extraction. If a premium/discount value marketing structure is implemented for these soybean seed quality components, producers will need information about how to produce soybean with high concentrations of these seed quality components. Previous investigations have shown that differences in seed oil and protein concentrations are inherent among cultivars (Simpson and Wilcox, 1983; Helms and Orf, 1998). But it has also been frequently observed that the same cultivar, when grown in different years or under different environments in the same year, varies significantly in seed composition. Significant genetic and environmental impacts on isoflavone concentration in soybean seed have also been reported. (Hoeck et al. 2000; Tsukamoto et al. 1995). However, further understanding of the relationships of the concentrations and yields of isoflavones, oil, and protein with seed yield of soybean is essential to soybean growers who may be given financial incentives to produce high-oil, high-protein, and (or) high-isoflavone soybean, and to soybean breeders for the selection of soybean cultivars that have genetic potential for high concentrations of oil, protein, and (or) isoflavones.

### OBJECTIVE

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To determine the relationships of the concentrations and yields of isoflavones, oil, and protein with seed yield of soybean simultaneously across a wide variety of production environments

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### **MATERIALS AND METHODS**

### Plant Material

•Soybean cultivars are presented in Table 1. The cultivars were selected to represent a range of maturity groups of soybean commonly grown in southwestern Ontario.

### **Field Experiments**

•Seed samples were collected from five field experiments conducted in Ontario which involved the determination of soybean responses to K fertilizer application and placement, soybean row width, tillage systems, etc.

•The five experiments will be referred to as Paris Direct, Kirkton Direct, Strathroy Direct, Paris Residual, and Kirkton Residual, respectively. In these experiment names, "direct" infers that K fertilization treatments were imposed directly to the soybean crop; while "residual" implies that K fertilizers were applied to the corn preceding the soybean crop, and no K fertilizer was applied after corn or during the soybean season. More details about the experimental treatments and management were presented in a previous publication (Yin and Vyn, 2005).

•When K was applied, the rate was 100 kg K ha<sup>-1</sup> as muriate of potash (0-0-50). Soybean row widths included 76, 38, and 19

•Soil-test K concentration is presented in Table 1.

### Seed Evaluation

•Seed yield was determined using a plot combine to harvest a central strip of soybean 1.0 m wide for the entire plot length from each plot, and adjusting the yield to moisture content of 130 g kg<sup>-1</sup>.

•Seed oil concentration was determined using GrainSpec (Foss Electric, Great Britain), near infrared reflectance spectroscopy calibrated with a gravimetric method. Seed protein concentration was measured using the same equipment as for seed oil calibrated by Kjeldahl (N × 6.25). Isoflavone concentration was determined using a high-performance liquid chromatography (HPLC) method modified from Franke et al. (1995). This method categorizes isoflavones into three aglycone groups of daidzein, genistein, and glycitein, which were summed to obtain total isoflavone concentration. The aglycone weight corresponded to approximately 55% of the weight in the naturally occurring glycosylated forms.

•The yields of oil, protein, daidzein, genistein, glycitein, and total isoflavone were defined as the products of seed yield and the seed concentrations of oil, protein, daidzein, genistein, glycitein, and total isoflavone, respectively, and were calculated for each experiment.

### **Statistical Analysis**

•Data of the concentrations and yields of each of these seed quality components were combined across all the five locations and two to three growing seasons before any statistical analysis in this study.

•In order to measure the differences in the concentrations and yields of these seed quality components among different seed yield levels, seed yield from each individual plot was grouped into low (<2.5 Mg ha<sup>-1</sup>), medium (2.5-3.0 Mg ha<sup>-1</sup>), high (3.0-3.5 Mg ha<sup>-1</sup>), and very high (>3.5 Mg ha<sup>-1</sup>) categories based on data distribution and common soybean yield standards. There were 87, 79, 77, and 61 individual plots for the low, medium, high, and very high categories, respectively.

•Analysis of variance was conducted for each of these seed quality components using the ANOVA procedure in the SAS package (SAS, 2002). Four seed yield categories were treated as the experimental treatments in this study. Mean separations were accomplished using Fisher's protected LSD test.

•Linear regression analysis was conducted using the REG procedure in the SAS package between the concentrations (or the yields) of each measured seed quality component and soybean seed yield on a plot basis. A total of 304 plots were used in all the regression analyses. Probability levels less than 0.05 were designated as significant for all analyses.

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Table 1. Initial soil K concentration (0-15 cm), soybean variety, yield mean, yield

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### **RESULTS AND DISCUSSION**

### **Oil, Protein, and Isoflavone Concentrations**

•Although oil concentrations in seed with high and very high seed yield categories were statistically lower than those under low and medium yield ratings (Fig. 1), the differences among the four yield categories were quite small. Protein concentration in seed did not differ significantly among the four yield categories (Fig. 1).

•Daidzein, genistein, and total isoflavone concentrations in seed increased significantly as seed yield increased from the low up to the high category (Fig. 2). Glycitein concentrations were significantly greater in the high and very high categories than in the low and medium yield ratings.

•The largest relative differences in daidzein, glycitein, genistein, and total isoflavone concentrations among the four yield categories were 67.0%, 17.7%, 39.6%, and 49.1%, respectively. This suggests that daidzein is the most variable and glycitein is most stable of the isoflavone components.

•Overall, on a concentration basis, both individual and total isoflavones varied with soybean yield level to a much greater magnitude than oil and protein (Fig. 1 and 2).

### Oil, Protein, and Isoflavone Yields

•Unlike oil and protein concentrations, both oil and protein yields showed a significant increase as seed yield increased from low up to very high (Fig. 3). The increases were 77% and 84%, respectively, for oil and protein yields when the seed yield category changed from low to very high. The increase in seed yield was the dominant contributing factor to the increases in oil and protein vields.

•Yields of individual and total isoflavones increased significantly as seed yield rating increased from low up to very high level (Fig. 4). The increases were 203%, 104%, 151%, and 169% for daidzein, glycitein, genistein, and total isoflavone yields, respectively, when seed yield category changed from low to very high.

### Regressions of Oil, Protein, and Isoflavone Concentrations with Seed Yield

•Although seed oil concentration was negatively and linearly related to seed yield when the data were combined across locations and years (Fig. 5), the decrease was quite small (about 4 g kg<sup>-1</sup>) when seed yield increased by over 1 Mg ha<sup>-1</sup>. The relationship between protein concentration and seed yield was not significant (Fig. 5).

•Concentrations of daidzein, glycitein, genistein, and total isoflavone were all positively and linearly related with seed yield (Fig. 6). The isoflavone concentration increases were much larger than the oil concentration increment when seed yield increased by

•Low R<sup>2</sup> values for these regression equations were probably due to the fact that only a single factor (seed yield) was used in these analyses. There are likely to be other factors contributing to the changes in these seed quality components.

### Regressions of Oil, Protein, and Isoflavone Yields with Seed Yield

•Unlike the variable constituent-specific responses for concentrations, the yields of oil, protein, daidzein, glycitein, genistein, and total isoflavone were significantly and positively related to seed yield (Fig. 7 & 8). However, individual and total isoflavone yields had greater relative increases than oil and protein yields.

### CONCLUSIONS

•Oil concentration in seed decreased very slowly, and protein concentration remained almost constant, as soybean yield

•Concentrations and yields of individual and total isoflavones, and yields of oil and protein, were all positively related to seed

•Daidzein was the most variable, and glycitein was the most stable isoflavone component.

•Our results suggest that even when soybean farmers plant cultivars that were not selected based on having high isoflavone concentrations, and even when seed oil and protein concentrations are little affected by yield level, high soybean seed yield can be accompanied by high seed isoflavone concentrations.

### ACKNOWLEDGMENTS

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### REFERENCES

•Caragay, A.B. 1992. Cancer-preventive foods and ingredients. Food Technol. 4:65-68.

• Franke, A. A., L. J. Custer, C. M. Cerna, and K. Narala. 1995. Rapid HPLC analysis of dietary phytoestrogens from legumes and from human urine. Proc. Soc. Exp. Biol. Med. 203:16-26. •Hartwig, E.E., and T.C. Kilen. 1991. Yield and composition of soybean seed from parents with different protein, similar yield. Crop Sci. 31:290-292.

- •Hasler, C. M. 1998. Scientific status summary on functional foods: Their role in disease prevention and health promotion. Food Technol. 52:63-70.
- •Helms, T.C., and J.H. Orf. 1998. Protein, oil, and yield of soybean lines selected for increased protein. Crop Sci. 38:707-711.
- •Hoeck, J. A., W. R. Fehr, P. A. Murphy, and G. A. Welke. 2000. Influence of genotype and environment on isoflavone contents of soybean. Crop Sci. 40:48-51.

•SAS Institute. 2002. The SAS System for Microsoft Windows. Release 8.2. Cary, NC. •Simpson, A.M., Jr., and J.R. Wilcox. 1983. Genetic and phenotypic associations of agronomic characteristics in four high protein soybean populations. Crop Sci. 23:1077-1081.

•Smith, K. 1991. Improvement of soybean composition to meet consumer/user demands. p. 71-78. In D. Wilkinson (ed.) Rep. of 21st Soybean Seed Research Conf. Chicago. Am. Seed Trade Assoc., Washington, DC.

•Tsukamoto, C., S. Shimada, K. Igita, S. Kudou, M. Kokubun, K. Okubo, and K. Kitamur. 1995. Factors affecting isoflavone content in soybean seeds: Changes in isoflavones, saponins, and composition of fatty acids at different temperatures during seed development. J. Agric. Food Chem. 43:1184-1192. •Yin, X., and T.J. Vyn. 2005. Relationships of isoflavone, oil, and protein in seed with yield of soybean. Agron. J. 97:1314-1321.