

## Evaluation of Several Agronomic Traits in ‘Essex’ By ‘Forrest’ Recombinant Inbred Line Population of Soybean [*Glycine max* (L.) Merr.]

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### Abstract

Crop yield is a polygenic complex trait and its improvement is a major goal of breeding programs. The objective of this study was to compare yield and its components along over a period of four years (2007-2010) in three locations in North Carolina using the ‘Essex’ by ‘Forrest’ recombinant inbred line (RIL) population of soybean (ExF, n=94). The RILs distribution for all traits showed higher means than their respective mid-parental values but do not differ significantly at  $P < 0.05$ . Nearly 45% of the lines germinated later than 6 days which is the mean for the slower germinating parent, Essex. In approximately 63%, the first flower appeared at 52.5 days, which is the mean mid-parental value. As for seed weight, RILs showed better performance than parental lines and 46% of the plants exceeded the higher yielding parent, Forrest. Seed weight showed the highest level of variation ranging from 54.1% for year to 70.7% for genotype. The lowest coefficients of variation (CVs) on average were calculated for flowering time and did not exceed 31.6%. In contrast, the year of the experiment caused the lowest level of variation for the traits studied while the genotype caused the highest level of variation. Seed germination was positively correlated with plant height ( $r = 0.441$  at  $P < 0.001$ ) and negatively correlated with both flowering time ( $r = -0.374$  at  $P < 0.001$ ) and seed weight ( $r = -0.357$  at  $P < 0.001$ ) across environments. Flowering time was found negatively correlated with plant height ( $r = -0.579$  at  $P < 0.001$ ) and positively correlated with seed weight. The ExF population performed well in all environments compared to other populations tested in

the same environments. The results presented here can be beneficial to NC soybean breeding programs that aim to create superior high yielding and disease free cultivars adapted to several NC environments.

**Key Words:** Soybean, seed yield, plant height, flowering time, days to germination.

### Introduction

Higher seed yield is a major goal of breeding programs in soybean [*Glycine max* (L.) Merr.]. Improved performance can be achieved through cultural practices and genetics. Successful plant selection requires knowledge and understanding of yield components, other important agronomic traits and their relationships. Nevertheless, yield is a quantitative characteristic which is difficult to be predicted and for this reason multi-year and multi-location field trials are needed (Yuan et al., 2002).

Study of quantitative traits is complicated due to the combined gene and environmental action. Environmental conditions, in particular, play an important role on gene expression and make breeding studies very challenging (Chenu et al., 2011). In general, phenotypic selection as a result of genotype by environment interactions is laborious for yield and other quantitative traits such as seed germination, flowering time or plant height. Yield is divided into many components that must be taken in consideration for the final selection. Seed weight is one of the most

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valuable components of yield, thus knowledge of interrelationship with other agronomic traits is crucial (Fehr, 1987). Seed germination is considered one of the most important traits for seed production and competitive yield performance (Kopich-Obusch and Diers 2006; Yari et al., 2010). Plant height was also an important characteristic in soybean evaluation and selection (Mansur et al., 1996; Orf et al., 1999; Yuan et al. 2002; Hyten et al. 2004). It is known that soybean plants with relatively higher height can have reduced yield potential due to lodging (Shapiro and Flowerday, 1987; SoyBase, 2011). In soybean, flowering time is closely related to maturity (Cober et al., 1996). Several genes have been identified that control these traits (Bonato and Vello, 1999; Cober and Voldeng, 2001; SoyBase, 2011) and they strongly interact with different environmental conditions (Cober et al., 1996). Early flowering time is a desirable trait because it often leads to higher yields.

Multi-location genetic studies for evaluating agronomic characteristics have provided valuable information to plant breeders and advanced selection process. Additionally, knowledge of correlations among important agronomic traits helps to making progress in conventional breeding procedures (Barnard et al., 2002). There are many studies in soybean that explored and reported interrelationships between several agronomic traits (Cicek et al., 2006; Oz et al., 2009; Mansur et al., 1996). Results for the relationship of plant height with seed yield are controversial. Cicek et al. (2006) showed that seed yield was strongly and positively correlated with plant height ( $r=0.58$  at  $P<0.001$ ). Yuan et al. (2002) and Oz et al. (2009) reported that these two traits not correlated significantly ( $r=0.018$ ). However, Ramteke et al. (2010) showed a negative and significant correlation between these characters ( $r=-0.447$  at  $P<0.01$ ). It is generally accepted that seed weight, as one of the components of crop yield potential, is positively correlated with seed yield (Smith and Camper, 1975). Correlation between flowering time was found positively correlated with seed germination ( $r=0.5$ ) by Watanabe et al. (2004), although the authors did not report the level of significance. Additionally, flowering time has been found to be positively correlated with plant height ( $r=0.401$  at  $P<0.01$ ) but negatively ( $r=-0.354$  at  $P<0.01$ ) with seed weight (Ramteke et al., 2010).

The objective of this study was to evaluate seed yield and other agronomic traits in different years (2007-2010) and locations in north Carolina using a recombinant inbred line (RIL) population of soybean between 'Essex' and 'Forrest' analyzed by Yuan et al., (2002) in southern Illinois.

## Materials and Methods

### Plant Material

In this study, we used the 'Essex' by 'Forrest' recombinant inbred line (RIL) population (ExF,  $n = 94$ ; Lightfoot et al., 2005). The population was provided to us by Dr. David Lightfoot of SIUC in 2007.

### Growth Conditions

Four seeds of 'Essex', 'Forrest', and each RIL were grown in pots of 15 cm x 14 cm (diameter x height) containing potting soil in two groups, the first, at the beginning of June 2007 and 2008 and the second in 2009 and 2010. The plants (4 per pot) were kept in the greenhouse at  $25\pm1^{\circ}\text{C}$  under natural daylight for 3 weeks. After 3 weeks, the two healthy plants among the four planted in the pots were transferred into the fields. The plants (2 per RIL) were tested in four different environments in North Carolina; Broadway in 2007 (BR07), Spring Lake in 2008 (SL08), and Fayetteville in 2009 and 2010 (FA09 and FA10). Fields BR07 [35.30N, 79.030W], SL08 [35.170N, -78.970W], in Harnett County, NC, and Fayetteville: and FA09 and FA10 [35.0N, 78.0W], Cumberland County, NC were free from obvious pathogen infestations.

The plants were kept in the field, watered with 2 liters of water every day until maturity of all RILs and the parents 'Essex' and 'Forrest' (112 days after planting, Lightfoot et al., 2005). No pesticides or herbicides were applied to the plants neither in the greenhouse nor in the field. The row-spacing and the spaces between individual plants in all rows were 30 cm which generated a 16 plants  $\text{m}^{-2}$ , in the field. A complete randomized block design with 2 replications was followed in planting. The soil type in all locations was sandy (rich in sand) – Carolina Sandhills.

### Trait Measurements

Several traits were measured in both groups of this population. The days to germination were recorded in the greenhouse while yield and its components (days to flowering, plant height, and seed weight) were recorded in the field. Plant height and seed weight were measured at harvest stage (R8).

### Statistical Analysis

Descriptive statistics and Shapiro-Wilk test for distribution normality were performed both for RI and parental lines at each location. Frequency distributions of RI lines were plotted for seed weight over locations. Direct comparisons between traits were based on calculated coefficients of variation (CV) for each variance component (year, location and genotype). Correlation coefficients between agronomic traits were calculated for each location for both years. Results of correlations between all independent variables are reported in a Pearson correlation matrix. All statistical analyses were performed on JMP 8.02 (SAS Institute Inc., Cary, NC, USA).

## Results

### Comparison of RILs and Parents

In Tables 1 the means, standard deviations, ranges and results of Shapiro-Wilk test of distribution normality for the agronomic traits measured for RILs were compared with parents across tested environments (BR07, SL08, FA09 and FA10). The RILs distribution for all traits showed means that were higher than their

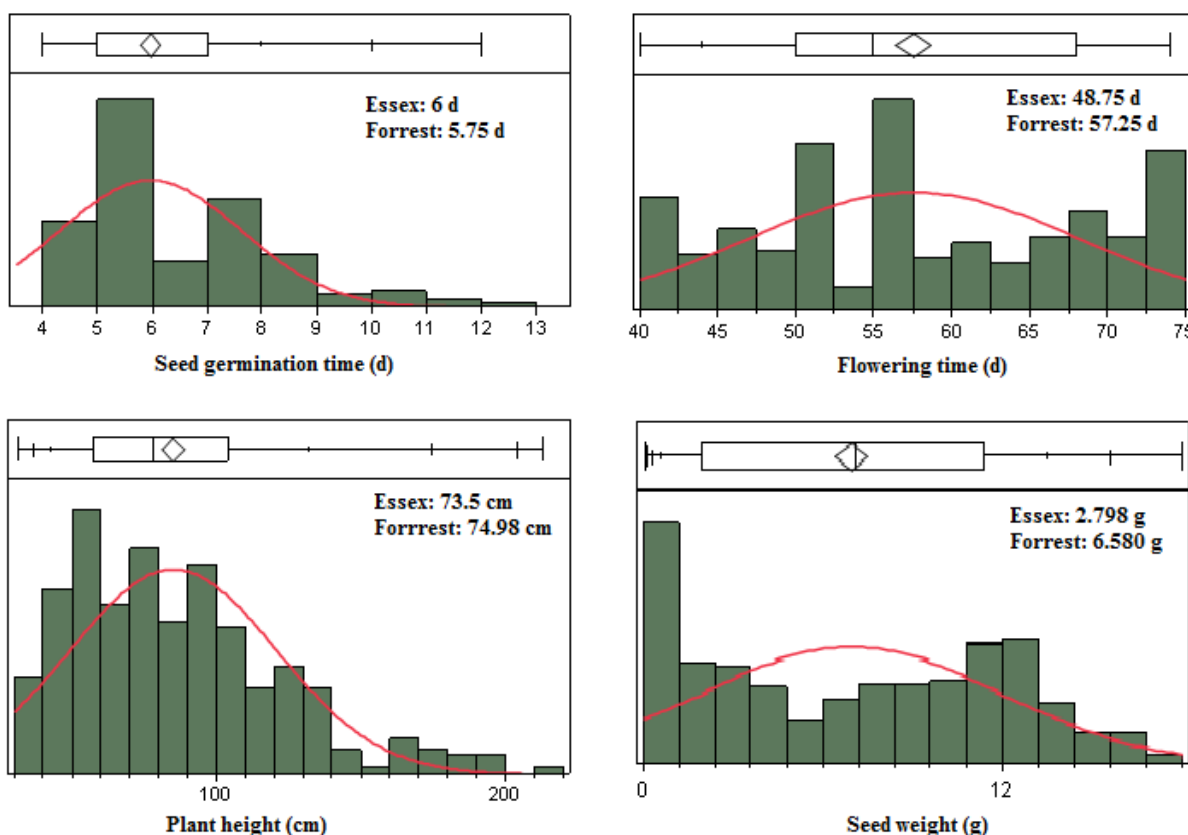
**Table 1.** Descriptive statistics and  $t$  tests results for differences between RI lines and midparental values for agronomic traits in a soybean recombinant inbred population from a cross between 'Essex' and 'Forrest' across testing environments (BR07, SL08, FA09 and FA10).

Trait	RIL		Essex		Forrest		Midparent		$t$ test	
	Mean	SD <sup>+</sup>	Range	P<W <sup>++</sup>	Mean	SD	Mean	SD	RI mean-Midparent	
<b>Seed Germination time (d)</b>	5.949	1.654	4-12	<.0001***	6.000	0.816	5.750	1.500	5.875	0.0736ns <sup>+++</sup>
<b>Flowering time (d)</b>	57.534	10.650	40-74	<.0001***	48.750	17.423	56.250	12.393	52.500	5.034ns
<b>Plant height (cm)</b>	84.842	35.611	31.2-213	<.0001***	73.500	26.221	74.975	42.722	74.238	10.604ns
<b>Seed weight (g)</b>	6.933	4.946	0.1-17.99	<.0001***	2.798	2.541	6.580	7.899	4.689	2.244ns

+Standard Deviation. ++ Shapiro-Wilk test for normality. +++ ns: not significant difference. \* Significant at  $P < 0.05$  probability level. \*\* Significant at  $P < 0.01$  probability level. \*\*\* Significant at  $P < 0.001$  probability level.

**Table 2.** Coefficients of variation for agronomic traits (seed germination time, flowering time, plant height and seed weight) in a soybean recombinant inbred population from a cross between 'Essex' and 'Forrest'.

	Seed germination time (d)	Flowering time (d)	Plant height (cm)	Seed weight (g)
<b>Year</b>	19.331	13.905	20.039	54.080
<b>Location</b>	20.793	14.462	27.993	55.964
<b>Genotype</b>	25.214	31.633	38.896	70.676



**Figure 1.** Frequency distributions of seed germination time (d), flowering time (d), plant height (cm), and seed weight (g) of 'Essex' x 'Forrest' recombinant inbred lines across testing environments (BR07, SL08, FA09 and FA10). Parental means are given for comparison.

**Table 3.** Correlation coefficients for agronomic traits in a soybean recombinant inbred population from a cross between 'Essex' and 'Forrest' and parental lines across testing environments (BR07, SL08, FA09 and FA10).

Trait	Flowering time (d)	Plant height (cm)	Seed weight (g)
Seed germination time (d)	-0.374***	0.441***	-0.357***
Flowering time (d)		-0.579***	0.237***
Plant height (cm)			-0.396***

\* Significant at  $P < 0.05$  probability level. \*\* Significant at  $P < 0.01$  probability level. \*\*\* Significant at  $P < 0.001$  probability level.

respective mid-parental values but do not differ significantly at  $P < 0.05$  (Table 1). The distribution of all traits deviated significantly from normality based on the results of Shapiro-Wilk test and showed positive skewness (Table 1). Nearly 45% of the lines germinated later than 6 days which was the mean for the slower germinating parent, 'Essex' (Fig. 1). In approximately 63%, the first flower appeared later than 52.5 days, which is the mean mid-parental value. RILs were on average 10 cm taller than the mean mid-parent value and almost half of them had a height of 75 cm or more (Fig. 1). As for seed weight, RILs showed better performance than parental lines and a 46% of the plants exceeded 'Forrest', which had higher yield potential than 'Essex' (Fig. 1) (Lightfoot et al., 2005). It is worth mentioning that the parents did not differ significantly for any of the studied traits.

#### Genetic Variation of Agronomic Traits

Coefficients of variation (CVs) were calculated for each traits and variance component (genotype, year and location) (Table 2). Between traits it was seed weight that showed the highest levels of variation that ranged from 54.1% for year to 70.7% for genotype. The lowest CVs on average were calculated for flowering time and did not exceed 31.6%. As for variance components the year of experimentation seemed to cause the less variation to the studied traits while the highest levels of variations occurred due to genotype.

#### Correlation Coefficients of Agronomic Traits

Correlation coefficients were estimated for each pair-wise combination of agronomic traits from the RILs at each location separately (Table 3). Overall, it was observed that although correlation coefficients were statistically significant only one was more than 0.5. More specifically, seed germination was positively correlated with plant height ( $r=0.441$  at  $P < 0.001$ ) and it was negatively correlated with both flowering time ( $r=-0.374$  at  $P < 0.001$ ) and seed weight ( $r=-0.357$  at  $P < 0.001$ ) across environments. Flowering time was found negatively correlated with plant height ( $r=-0.579$  at  $P < 0.001$ ) and positively correlated with seed weight. The latter was pretty low given that coefficient did not exceed 23.7% but it was highly significant at  $P < 0.001$ . Although, plant height was moderately and negatively correlated with seed weight ( $r=-0.396$ ), it was statistically significant at  $P < 0.001$ .

#### Discussion

The study of agronomic traits and seed weight of a RIL population between two soybean lines, 'Essex' and 'Forrest' was presented in this study. Analysis of the distribution revealed that it was continuous but deviated significantly from normality for all traits studied here (Table 1). The population means across the tested environments were not significantly higher than mid-parental average but they were significantly higher in seed weight than low yield parent, 'Essex'. Genotypic variation was high for all traits but especially for seed weight with values that exceeded 54% (Table 2). The observed variation in the RIL population can be beneficial and has practical applications for improvement programs. Correlations among traits in the RILs provide a better understanding of the population (Table 3). Flowering time was negatively correlated with both seed germination and plant height; moderately with the former and highly with the latter. Seed germination and flowering time are strongly related to environmental conditions. In Arabidopsis, Chiang et al. (2009) reported that there is a major gene, FLC, which controls flowering and also promotes temperature-controlled seed germination. These traits are probably closely related in soybean and are regulated mainly by seasonal conditions. Environmental conditions and specifically photoperiod seems to control post-flowering development too (Grimm et al., 1994; Steward et al., 2003). This must be a reasonable explanation to the fact that the results of this study are not in accordance with a previous study which suggested that early flowering boosts the development of the reproductive system ceasing vegetative growth and as a result the plants are shorter in height (Panthee et al., 2007).

The top priority of a breeding program is yield. It is generally accepted that when agronomic traits are found significantly correlated with seed yield, this can be helpful in selecting desired genes for particular character. This study indicated that seed yield is negatively correlated with germination time and plant height. Faster seed emergence leads to improved yield because of reduced pest damage and increased drought tolerance of the seedlings (Mussa et al., 1999; Harris et al., 2000). Also, shorter plants tend to be more resistant to lodging which reduces yield up to 10%. Panthee et al., 2007 reported that there is a significantly positive correlation between plant height and lodging while the correlation between lodging and yield is significantly negative. However, it should be mentioned that correlation coefficient that are low ( $r < 0.5$ ), even if they are statistically significant, cannot be helpful in efficiently selecting for high

yield using agronomic traits. Similarly, seed linoleate, linolenate, and oil contents were negatively correlated with flowering and maturity dates (Bashlava et al., 2008).

The ExF RIL population performed relatively well in all NC environments it was tested in (Broadway, Spring Lake, and Fayetteville) compared to 'Hartwig' by 'Flyer' RIL population (HxF, n=92; Kazi et al., 2008) grown in Spring Lake in 2010 (Ouertani et al., 2011). Several other studies are underway to develop superior soybean cultivars with high yield, glyphosate resistance, soybean cyst nematode (SCN) resistance, sulfonyl urea herbicide (STS) resistance, and other agronomic traits in NC environments (Mavromatis et al., 2002; NC Soybean Producers Association, 2010; Bashlava et al., 2008) and our results can be beneficial to these NC soybean breeding efforts.

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