

TRANSGENE EFFECT ON THE STABILITY OF COTTON CULTIVARS IN LOUISIANA

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Abstract

Studies were conducted in 2001 to determine the effect of transgenes on mean performance and stability of selected cotton cultivars for a variety of non-target traits. Multi-environment trials are conducted every year and mean performance is often calculated as an average of cultivar performance over years and locations. However, comparisons of mean performance are not sufficient for cultivar evaluation without an understanding of the causes of genotype x environment (GE) interactions. A graphical data summary called GGE Biplot can be used to characterize GE interactions and make meaningful comparisons between cultivars. A multi-environment data set was used with GGE Biplot to compare conventional and transgenic cotton cultivars. Preliminary analyses of variance (ANOVA) in SAS revealed that GE interactions were present only for plant height, number of nodes, fuzzy seed index, and lint percent. There were no GE interactions for yield. Plant height at 60 DAP, fuzzy seed index, and lint percent were analyzed in GGE Biplot, and differences, based on environment, between conventional and transgenic cultivars were further examined. Transgenic and conventional cultivars did not behave similarly for plant height, fuzzy seed index, or lint percent at the four locations in this study. The relative differences between transgenic plants and their conventional counterpart differed based on environment and crossover interactions were present. Additionally, transgenic plants were taller, and their seed were heavier than conventional plants, although the degree of which these differences were present was dependent on location. In all cases, conventional plants had higher lint percentages than transgenic plants.

Introduction

Cotton is an indeterminate perennial crop grown as an annual in the United States. Recently, the introduction of a number of transgenic technologies has yielded many transgenic cotton cultivars. Multiple herbicide and insect resistant cultivars have been commercialized and there are several varietal options utilizing either a conventional cultivar, or an herbicide resistant or insect resistant cultivar similar to a high-yielding conventional recurrent parent. Each transgenic line results from a separate insertion event, is usually stable, and segregates with Mendelian expectations (Kohel, 2000). Transgenic cultivars have been developed by backcrossing a transgenic line into a high-yielding conventional cultivar until the transgenic line is near-isogenic with the conventional recurrent parent (Bowman et al., 2000; Robinson and McCall, 2001).

The performance of a cotton cultivar is dependent on the genetic capacity of the cultivar, the environment where the cultivar is grown, and the interaction between the cultivar and the environment (Kerby et al., 2000; Myers, 1997; Yan, 2001a,b). The term genotype x environment (GE) interaction refers to variation that cannot be explained by the genotype main effect or the environment main effect (Yan, 2001b). The environment, or location, refers to all biotic and abiotic factors that can influence plant growth at that location including weather (temperature, wind, precipitation, heat, cold, drought), impact of planting date, plant stand, disease pressure, soil type, and management factors including items such as irrigation, fertility, use of plant growth regulators, weed control pressure and practices, insect pressure and control, etc. (Kerby et al., 2000). When the GE interaction is significant, comparisons based strictly upon the mean may not be adequate (McPherson and Gwathmey, 1996). Useful comparisons between conventional and transgenic cotton cultivars should be made based on mean performance of a cultivar (genotype main effect) and the stability of that cultivar (GE interaction) over a range of pertinent environments.

Recently, much variability in the performance of cotton cultivars has been attributed to differences in environment (Kerby et al., 2001); however, for some response variables, genes have a greater effect than the environment (Meredith and Bridge, 1972). Meredith and Bridge (1972) reported that, within upland cotton genotypes, there is little non-additive gene action in fiber length, strength, and fineness, indicating that genes determine those fiber properties. Other studies have suggested that the relative genetic and environmental influences on fiber strength are determined by a few major genes, rather than by variations in the growth environment (May, 1999). However, large differences in environment can inhibit cotton genotypes from reaching their full genetic potential (Green and Culp, 1990). Many environmental factors affect cotton performance; specifically, cotton canopy architecture, plant height, and branch formation can be affected by temperature (Hanson et al., 1956; Hodges et al., 1993; Reddy et al., 1990), growth-regulator application (Cadena and Cothren, 1996; Reddy et al., 1990), light intensity (Hanson et al., 1956; Sassenrath-Cole, 1995), and herbivory by insects and other animals (Rosenthal and Kotanen, 1994; Terry 1992). Bradow et al. (1997a,b) showed that boll retention, an important yield characteristic, was affected by macro-environment, where the macro-environment was irrigation method. Variability even exists within characteristics. Gipson and Joham (1969) showed that early-stage fiber elongation, which is controlled more by genes than

environment, was highly temperature dependent and late-fiber elongation was temperature independent, showing that environmental variation can influence traits controlled primarily by genotype. When genotypic effects respond differentially according to different environments, a GE interaction is present (Kerby et al., 2001).

Numerous methods for analyzing varietal differences based on their consistency in response to environments have been developed (Lin and Binns, 1988; Kang and Pham, 1991; Pinthus, 1973). One strategy involves factorial regression of the GE matrix against environmental factors, genotypic traits, or combinations thereof (Baril et al., 1995). A second strategy, the additive main effects and multiplicative model (AMMI), involves correlation or regression analysis that relates the genotypic and environmental scores derived from principal component analysis of the GE interaction matrix to genotypic and environmental covariates (Zobel et al., 1988). GGE Biplot performs analyses similar to the popular AMMI model, but focuses on the G and GE components relevant to cultivar evaluation.

The objective of this research was to compare the stability of conventional cotton cultivars and their near-isogenic transgenic derivatives. In doing so, we sought to identify differences between conventional and transgenic cotton cultivars, the traits that are affected, and the environments that influence variation. Several non-target traits were evaluated and GGE Biplot was used to compare the stability and performance of cotton cultivars in four environments in Louisiana. Similarities or differences between near-isogenic lines were determined and a measure of stability was used to examine GE interactions over the four locations.

Materials and Methods

Cotton cultivars were planted at the Red River Research Station in Bossier City, the Dean Lee Research Station in Alexandria, and the Northeast Louisiana Research Station in St. Joseph and Winnsboro, Louisiana in 2001. These four test locations differed in weather patterns, soil type, geographic location, and management practices and provide a representation of the major cotton-growing areas of Louisiana. Cotton management decisions differed at all four locations to achieve optimal growth and necessary inputs throughout the growing season.

A randomized complete block design was used and four-row plots (4 x 15 m) were replicated three times for each cultivar at each location. Cultivars included in this study were Stoneville (STV) 474, STV 4691B, STV 4793R, STV 4892BR, STV BXN47, Suregrow (SG) 501, SG 501BR, and Deltapine (DP) 5415, DP 5415RR, and DP NuCOTN 33B. Data were collected at 60 and 90 days after planting (DAP) and at harvest. Yield, yield components and fiber quality were calculated at or after harvest. Cotton fiber quality measurements were obtained from the Cotton Fiber Testing Laboratory in Baton Rouge, Louisiana.

In-season data collected 60 and 90 DAP and at harvest included plant height, number of nodes, SPAD readings, vegetative weight and reproductive weight. Yield components included boll weight, lint weight, fuzzy seed index, seed index of acid-delinted seed, and yield. Fiber properties analyzed were fiber micronaire, strength, length, uniformity and elongation.

All response variables were subjected to an analysis of variance (ANOVA) in SAS to determine the presence or absence of GE interactions. Response variables that were significant for GE interactions were analyzed in GGE Biplot and stability and mean performance were characterized. Similar to the widely-accepted AMMI model, GGE Biplot is a recently released Windows-based software package designed to examine genotype (G) main effects and GE interactions using rank-two matrix multiplication and singular value decomposition (Yan, 2000; Yan, 2001a; Yan and Hunt, 2001b).

Results and Discussion

Analysis of Variance

Results for the analyses of variance are given in Table 1, which indicates the relative magnitudes of G, E, and GE interaction. Location was an important source of variation for plant height and accounted for 88% of the total amount of variation based on the Sums of Squares (SS). Other studies have shown that location typically accounts for >80% of total variation (Kerby et al., 2001). This is not surprising considering the large effect that location has on plant growth and morphology. However, location only accounted for 23 and 22% of the total SS for fuzzy seed index and lint percent, respectively. Genotype and GE interactions were primarily responsible for the highest variation in these two parameters. For lint percent, the large GE interaction, relative to G and E, suggests the possible existence of a discriminating, or different, environment.

Plant Height

Figure 1, or the average environment coordinate (AEC) biplot, is a two-dimensional graphical representation of a multi-environment data set with principle component (PC) 1 and PC2, which are unit-less measures, on the x- and y-axis, respectively. For this data set, PC1 and PC2 accounted for 94% of the total variation in G and GE, which suggests that this biplot is a good approximation of mean performance and stability (Figure 1). The average environment, which is generated

by the biplot and denoted by the red circle near Winnsboro (WINN), is bisected by a red line that passes through the biplot origin. The direction of the arrow indicates higher values for the parameter measured, in this case plant height, such that moving along the line in the direction of the arrow means taller plants and away from the arrow indicates shorter plants. In this manner, mean performance can be evaluated. The ten light-gray lines are unit-less measures and exist only to rank or evaluate the cultivars. The dark blue line, which runs perpendicular to the AEC axis and also passes through the biplot origin, indicates stability of any given cultivar. Distance, either up or down from the AEC axis and parallel to the blue line, is a measure of cultivar stability such that further distances from the AEC axis indicate less stability, and cultivars clustered near or on the AEC axis are highly stable. Figure 1 suggests that the two least stable cultivars, SG 501BR and STV BXN47, are also very tall plants, and that their conventional recurrent parents are much more stable, although they are shorter. This indicates that SG 501BR and STV BXN47 performed very well in some environment, but performed erratically over these four locations. In fact, SG 501BR, which is located near SJOE, was the tallest plant in St. Joseph (SJOE), but not the tallest in Alexandria (ALEX), Bossier City (BCITY), or WINN, hence the instability of this cultivar. In SJOE, SG 501BR might have higher seedling vigor or growth rate than at any other location. Averaged over all environments, it was the third tallest cultivar, but it would not hold that ranking if SJOE were not in the analysis, or the range of environments. A similar situation exists for STV BXN47, which was the tallest cultivar in ALEX, but one of the shortest in SJOE. In contrast to the instability of STV BXN47, the later Stoneville transgenics, STV 4691B, 4793R, and 4892BR, were more stable than the conventional parent, STV 474, and much more stable than STV BXN47 (Figure 1). All transgenic cultivars were taller than their conventional parent averaged over location, regardless of the level of stability. Figure 2 is a graphical comparison between STV 474 and STV BXN47. The two cultivars to be compared are contained in red circles. The dark blue line, which passes through the biplot origin and is perpendicular to the red dotted line connecting the two cultivars, is called the equality line. It represents equality between the two cultivars such that they should grow equally tall in any virtual environment that lies on the equality line. Cultivars have higher values in those environments on the same side of the line and lower values in environments located on the other side of the line. STV BXN47 is much taller in ALEX and BCITY, somewhat taller in WINN, and shorter in SJOE than STV 474, hence the GE interaction (Figure 2).

Fuzzy Seed Index

Location, genotype, and GE interactions accounted for 23, 59, and 18%, respectively, of the total variation in SS (Table 1). This suggests that fuzzy seed index is controlled more by genotype than by location and that these genotypes are responsive to locations in this study. The AEC graph for fuzzy seed index shows that there is little variability between the later STV transgenic plants, STV 4691B, 4793R, and 4892BR (Figure 3). In addition to their close proximity to each other, they are all very stable (close to the AEC axis) and their seed were very heavy, particularly in SJOE. STV BXN47 seed weights were more variable than the later STV transgenics and the STV 474 conventional parent (Figure 3). SG 501 and SG 501BR were both highly stable, but the transgenic SG 501BR had heavier seed weights than the conventional parent. In all cases, transgenic seed were heavier than the seed of the conventional recurrent parent (Figure 3). Comparisons between STV 474 and STV 4691B indicate that environment influences the relative differences in seed weight between these two varieties (Figure 4). STV 4691B had heavier seed weights in WINN, ALEX and SJOE; however, something in BCITY reduced the height advantage that STV 4691B had over the conventional, recurrent parent STV 474 (Figure 4).

Lint Percent

For lint percent, 22, 38, and 40% of the total SS were attributed to location, genotype, and GE interactions, respectively, (Table 1). As evidenced by the 40% of total variation due to GE interactions, it is clear that lint percent is more responsive to GE interactions than any other trait in this study. Also, location accounted for less of the total variation in lint percent, which was more heavily influenced by GE interactions and genotype than any other trait. Studies conducted by Kerby et al. (2001) demonstrated that location accounted for varying amounts of the total variation in cotton fiber properties. In their study, location accounted for 85, 59, 29, and 55% of the total variation in SS for fiber length, micronaire, strength, and lint percent, respectively. Other studies have shown that, in general, fiber micronaire, length, and strength were lower in transgenic cultivars than in conventional cultivars (Moser et al., 2001). Conventional cultivars had higher values for lint percent than their respective transgenic derivatives (Figure 5). There is a greater discrepancy between lint percent of SG 501 and SG 501BR than there is between STV 474 and any STV transgenic derivatives (Figure 5). This could imply that the STV 474 conventional parent more effectively compensates for smaller seed and shorter plants than the SG 501 conventional parent. Several examples of compensatory shift have been documented in the literature. Moser et al. (2001) found that DP 5415RR, a transgenic cultivar, produced fewer bolls per acre than the conventional recurrent parent DP 5415, but each boll contained more lint per boll, which resulted in very similar yields.

Conclusions

GGE Biplot can effectively examine multi-environment trial data sets and provides a means of cultivar evaluation utilizing both mean performance and stability of cultivars. It is clear that non-target traits can be affected by transgene insertion and that the differences between conventional and transgenic cultivars are affected according to the environment in which they

are grown. Some traits are more likely to be affected by environmental variation than others; yield, which is not a highly-heritable trait, was not affected by GE interactions (data not shown). Other studies have found that yield did not differ between conventional and transgenic varieties (Robinson and McCall, 2001; Moser et al., 2001). This could be due to the fact that transgene lines are selected for similarities to the recurrent parent and for yield, with yield being the most important characteristic. Though transgenic plants are made to contain a high percentage of the recurrent parent, absolute similarity might never be attained; therefore, differences between conventional and transgenic cultivars should be expected, but rigorous selection for yield can equalize those differences. Even though transgenic plants grew taller and had heavier seed, conventional parents had higher lint percents and all cultivars yielded equally over these four environments.

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Table 1. Genotype (G), environment (E), and genotype x environment (GE) variance terms for plant height, fuzzy seed index, and lint percent.

Source	Plant Height			Fuzzy Seed Index			Lint Percent		
	df	SS	Pr>F	df	SS	Pr>F	df	SS	Pr>F
E	3	943	<.0001	3	16	<.0001	3	213	0.0005
G	8	49	0.0007	8	41	<.0001	8	369	0.0003
G x E	23	85	0.0019	23	13	0.0400	23	387	0.0768

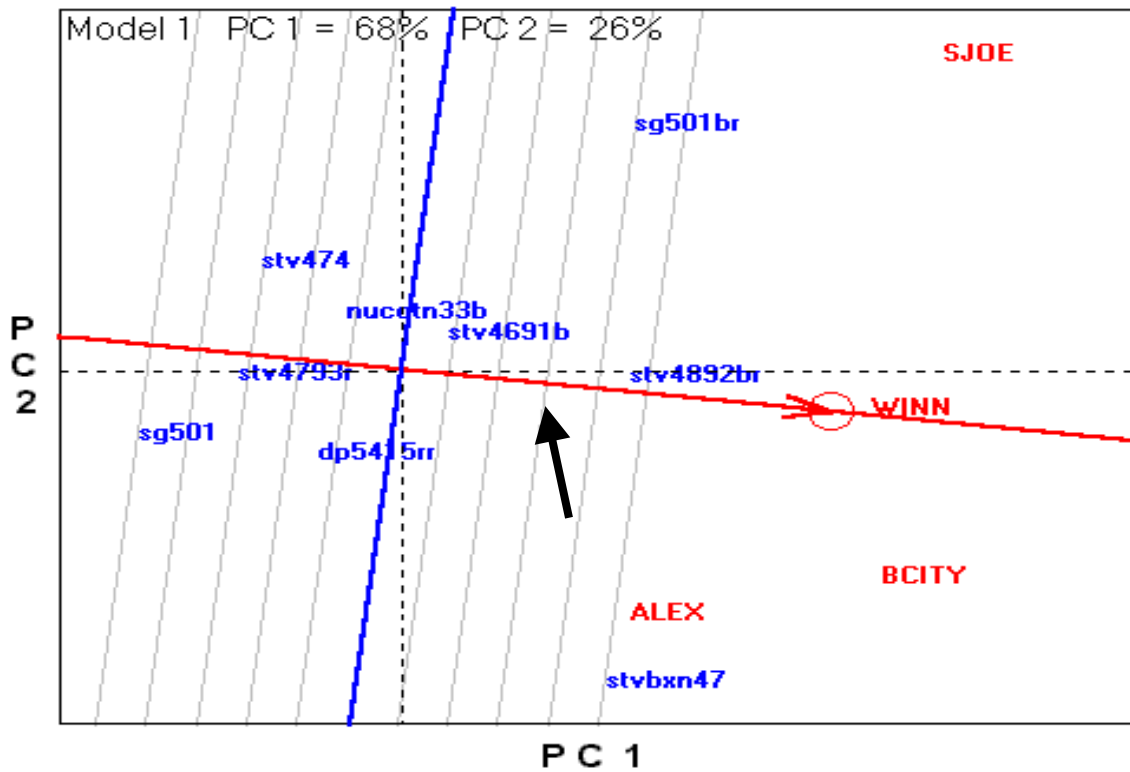


Figure 1. Two-dimensional biplot showing mean height and stability of nine cultivars at 60 DAP over four environments.

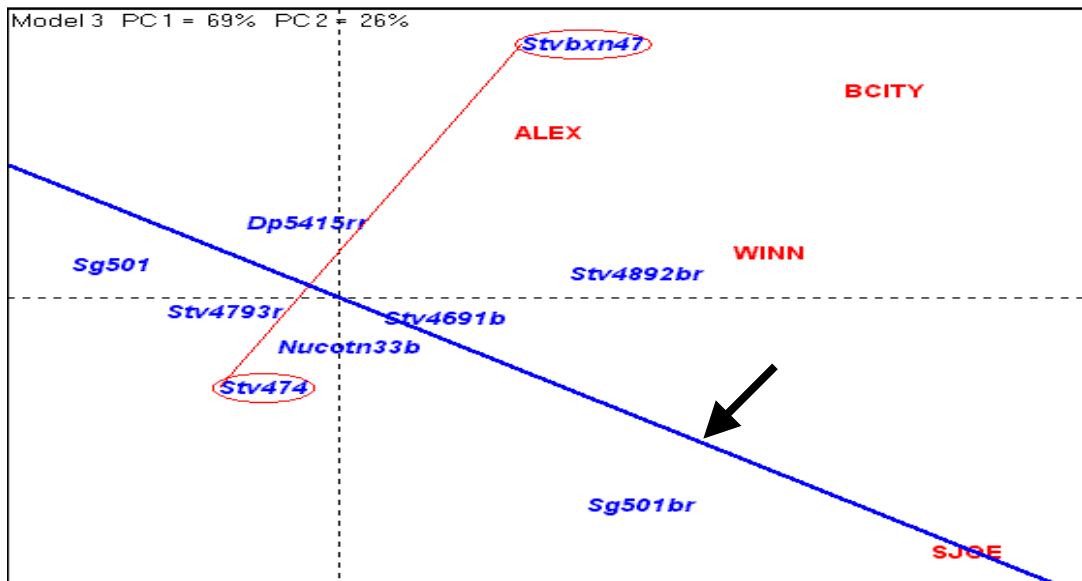


Figure 2. Biplot comparing plant heights of STV 474 and STV BXN47 at 60 DAP over four environments.

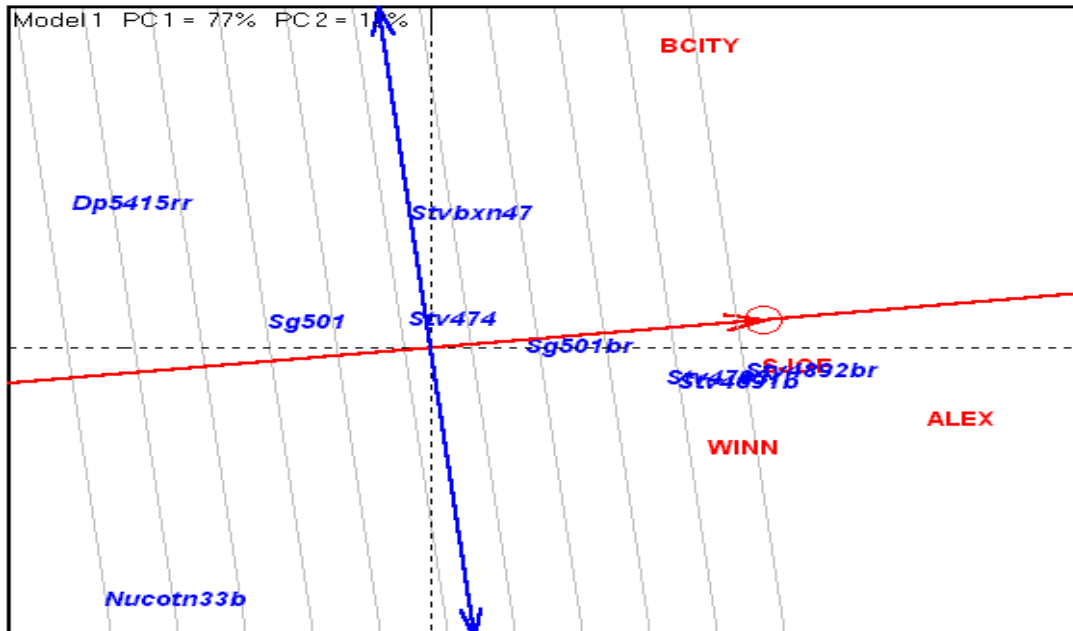


Figure 3. Two-dimensional biplot showing mean fuzzy seed index weight and stability of nine cultivars over four environments.

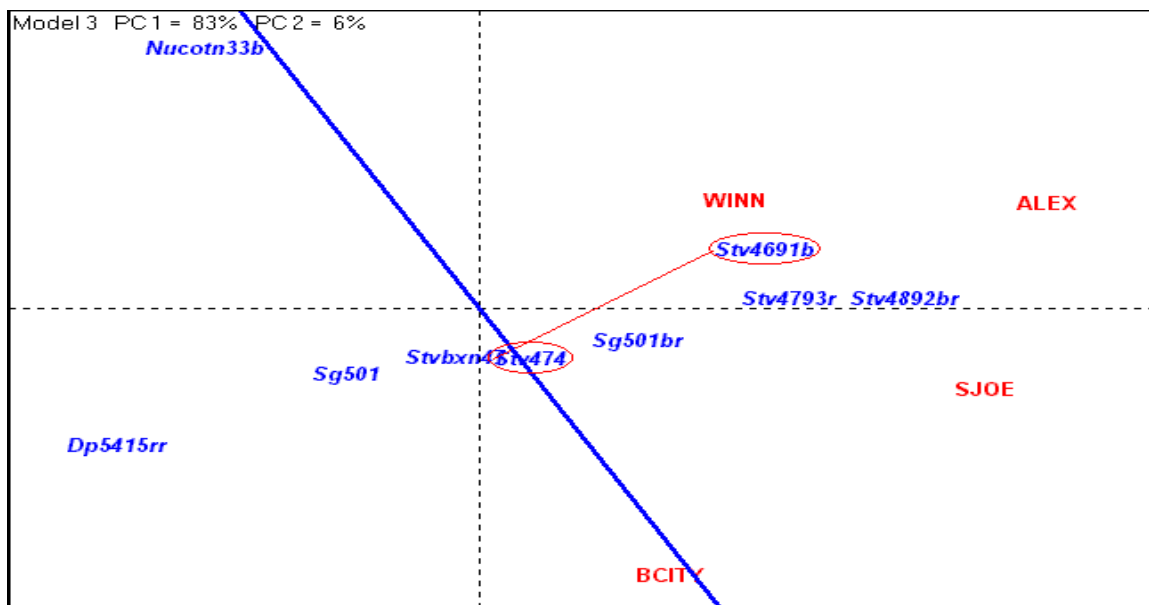


Figure 4. Biplot comparing fuzzy seed index of STV 474 and STV 4691B over four environments.

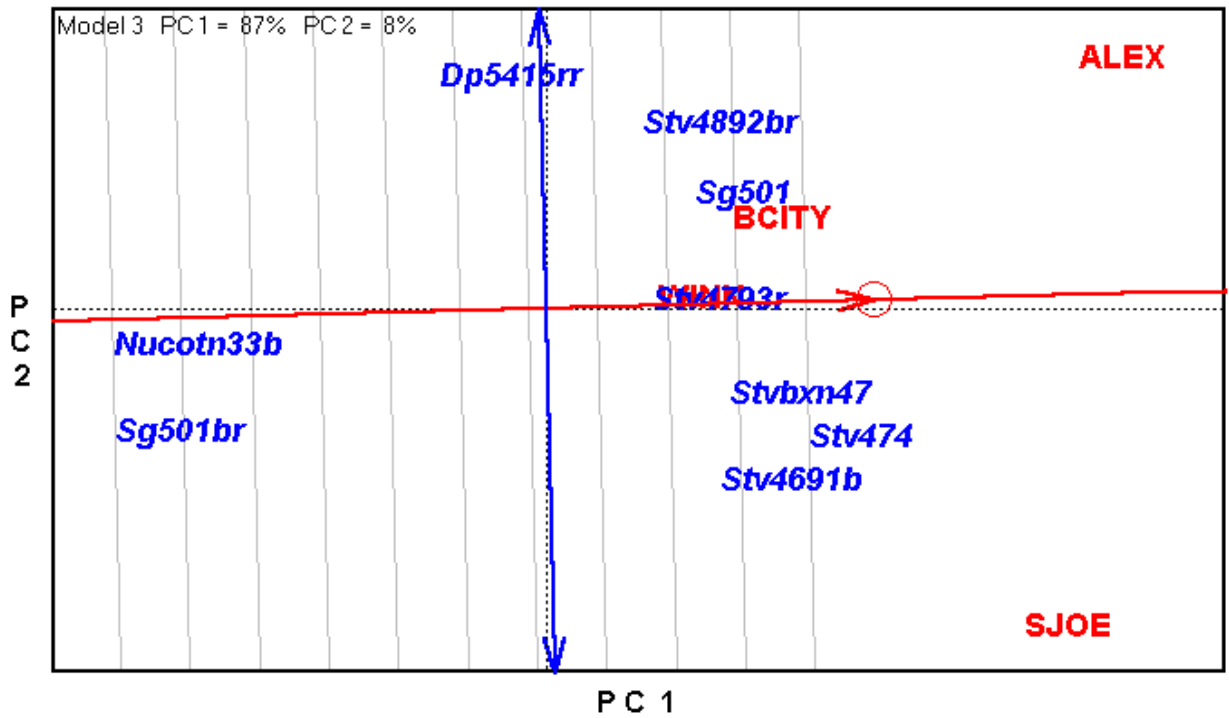


Figure 5. Two-dimensional biplot showing mean lint percent and stability of nine cultivars over four environments.