RESPONSE TO AFLATOXIN AND GRAIN COMPOSITION IN EXOTIC MAIZE GERMPLASM

A Thesis

by

REBECCA JOANN CORN

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

August 2007

Major Subject: Plant Breeding

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Co-Chairs of Committee,	Javier Betran
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ABSTRACT

Response to Aflatoxin and Grain Composition

in Exotic Maize Germplasm. (August 2007)

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Exotic germplasm has potential to provide new alleles for disease and insect resistance. US maize (*Zea mays* L.) currently lacks genetic resistance to *Aspergillus flavus*, a fungal pathogen that produces aflatoxin in maize kernels. Aflatoxin is one of the main limitations to maize production in hot, dry regions like the Southern US because of the harmful effects on humans and animals and subsequent marketing regulations. Two experiments were conducted to evaluate different exotic maize collections for response to aflatoxin. Exotic adapted maize lines, known as LAMA lines, were found to accumulate less aflatoxin than US hybrids in tests across Southern Texas. Exotic introgression lines developed by The International Center for Maize and Wheat Improvement (CIMMYT) including inbred lines, yellow hybrids, and white hybrids, were more resistant to aflatoxin than US inbred lines and hybrids in field trials in Texas, Georgia, and Mississippi.

Another experiment evaluated the grain composition of hybrids with exotic adapted LAMA maize lines and a collection of US hybrids, quality protein maize (QPM) hybrids, and advanced breeding lines using near-infrared spectroscopy. Individual

LAMA lines and advanced breeding lines have higher starch content than US hybrid checks. Starch content was the primary grain composition trait of interest as an enhanced-value market has emerged for high starch maize hybrids. Limited germplasm has been analyzed for grain composition because wet chemistry analysis methods required large sample sizes and were time and labor intensive. The near infrared spectroscopy (NIR) method requires a relatively small sample and is a non-destructive analysis method. In this study, NIR was effective at ranking genotypes based on starch, oil, and protein content of the grain.

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These projects were collaborative efforts, and I would like to thank CIMMYT for providing material and USDA in Georgia and Mississippi for evaluating the CIMMYT material at their locations. I would also like to thank former grad students and everyone involved in developing the LAMA material.

Special thanks to Kerry Mayfield who has helped me through all phases of this project, and to all of my fellow grad students and the student workers who have been great to work with. I've really enjoyed this experience.

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CHAPTER I

INTRODUCTION

Maize (*Zea mays* L.) is an important crop worldwide used for food, feed, and industrial purposes. In the United States, the majority of maize production is in the Midwest, but there is a substantial amount of maize grown in the southern US. In 2004, 1,680,000 acres were harvested in Texas at an average yield of 139 bushels per acre for a total crop value of $595,476,000 (NASS, 2005).

Genetic diversity in maize became a prominent issue when southern leaf blight devastated the maize crop. In the late 1960's, little genetic diversity existed in maize because US hybrids extensively used germplasm with Texas male-sterile cytoplasm. That cytoplasm was susceptible to the Race T pathotype of *Bipolaris maydis*, the causal organism of southern leaf blight in maize (Smith, 1988).

A narrow germplasm base is not unique to the US or to maize. Agricultural systems in all developed countries tend to be characterized by widespread cultivation of crop varieties from a relatively narrow and uniform germplasm base (Smith, 1988; Simmonds, 1993). A decrease in genetic diversity usually accompanies crop improvements achieved by plant breeding (Stuber, 1978). The US germplasm base continues to narrow as breeders increasingly rely on crosses among elite lines (Goodman, 1992). Maize breeders contribute to narrow genetic diversity by focusing on short-term goals (Betran et al., 2005). To increase genetic diversity, breeders can use

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This thesis follows the style of Crop Science.

tropical and subtropical maize collections as a source of new genes (Echandi and Hallauer, 1996). Genetic diversity in US maize is primarily limited to the corn belt dent race out of about 300 races worldwide (Brown, 1975). Six racial groups, less than five percent of all available sources, account for most commercial maize worldwide (Goodman, 1985). In the US, most commercial hybrids are derived from just six to eight inbred lines (Goodman et al., 1988, 1992; Smith et al., 1999; Tallury and Goodman, 1999; Betran et al., 2005). While many US hybrids derive some of their germplasm from those six high-performance inbreds, the other parts of these hybrids come from diverse origins so the germplasm base is not as narrow as some people suggest (Duvick, 1975, 1981).

The US maize germplasm base includes 3% tropical germplasm (Goodman, 1985, 1992; Goodman et al. 1988; Smith et al., 1999). The amount of tropical germplasm in US hybrids remains small, but is gradually increasing (Duvick, 1981, 1984; Goodman, 1985, Betran et al., 2005). Seed companies surveyed in 1981 predicted exotic germplasm incorporated into Corn Belt hybrids would increase to five to ten percent within 50 years (Goodman, 1985).

The primary reasons to use exotic germplasm are:

1. Increased genetic diversity provides a safeguard against unpredictable biological and environmental hazards (Stuber, 1978; Geadelmann, 1984; Albrecht et al., 1987; Goodman et al., 1988, 1992; Michelini and Hallauer, 1993; Tallury and Goodman, 1999; Betran et al., 2005).

2. Exotic germplasm is a source of genes for specific traits including aflatoxin resistance, drought tolerance, and husk coverage (Geadelmann, 1984; Albrecht et al., 1987; Betran et al., 2005).
3. Exotic germplasm is a source of favorable alleles for increased yield and enhanced heterosis (Geadelmann, 1984; Albrecht et al., 1987; Goodman et al., 1988; Tallury and Goodman, 1999).
4. Increased genetic diversity leads to increased flexibility in seeking efficient alternative uses for the crop (Goodman et al., 1988). Alternative uses of maize include ethanol, industrial starches, and biodegradable starches.

Plant breeders need genetic variability to make progress through selection so an increase in useful diversity in the germplasm base is always desirable. Breeders and geneticists also generally agree that decreased genetic variability is accompanied by increased genetic vulnerability (Brown, 1975). Exotic germplasm, especially tropical, is often suggested as a source of genetic diversity to widen the germplasm base in US maize (Goodman, 1992).

Disease and insect resistance are the most common traits maize breeders seek in exotic germplasm, but the US germplasm doesn't have a monopoly on high-yield genes (Goodman et al., 1990; Goodman, 1992; Tallury and Goodman, 1999). Inbred lines containing tropical germplasm are competitive in crosses with temperate lines (Tallury and Goodman, 1999; Lewis et al., 2003).

While most breeders agree that exotic germplasm is a source of desirable traits, several challenges limit their use. Tropical inbreds are poorly adapted to temperate

environments, and often express several of the following characteristics: excessive lodging, poor cold tolerance, poor floral synchronization, disease susceptibility, late maturity, high grain moisture and slow dry down, barrenness in high plant densities, photoperiod response, tall plants, large tassels, and high ear placement (Goodman, 1985; Goodman et al., 1990; Uhr and Goodman,1995; Echandi and Hallauer, 1996; Tallury and Goodman, 1999; Lewis et al., 2003).

Genetic linkages between favorable and unfavorable genes are difficult to break and pose another challenge to breeders working with exotic germplasm (Geadelmann, 1984; Goodman, 1985; Echandi and Hallauer, 1996). Exotic germplasm has not undergone as many cycles of selection as elite adapted material so the genetic component of exotic populations is 40 years behind current temperate breeding materials (Goodman, 1985; Echandi and Hallauer, 1996). Use of exotic germplasm is also limited by a lack of information about how exotic germplasm combines with adapted germplasm and the resulting loss of heterotic patterns (Goodman et al., 1988; Echandi and Hallauer, 1996). A lack of information also limits selection of the best exotic lines to use in a temperate breeding program. There is currently no basis to choose among collections of exotic material so often the choice of exotic lines has been rather random (Stuber, 1978; Goodman, 1985; Echandi and Hallauer, 1996).

Incorporating exotic germplasm into hybrids extends the amount of time that is required to develop a commercial maize hybrid. Twenty five to 30 years could be required for classification into US heterotic groups, adaptation, and prebreeding of unadapted exotic germplasm (Smith et al., 1999). Maize breeders rely on germplasm of

proven usefulness when they are under constant pressure to develop new cultivars (Brown, 1975).

The use of exotic germplasm is also limited by the fact that useful genetic variation still exits in elite Corn Belt germplasm (Brown, 1975; Duvick, 1981; Geadelmann, 1984). Since useful variation exists in adapted material, breeders will focus on it until we gain more information about how to identify useful genes in exotic materials and how to transfer them efficiently into elite adapted material because we do not need diversity of deleterious genes (Duvick, 1981).

The goal of this study is to screen exotic maize lines for traits lacking in US maize germplasm. This study includes evaluating the response of exotic maize lines to *Aspergillus flavus* and their starch, oil, and protein content compared to other germplasm collections. Two collections of exotic material were studied, the LAMA lines, a collection of exotic early generation lines that were developed by the Texas Agriculture Extension Service and adapted to Texas, and a collection of inbred lines, yellow hybrids, and white hybrids developed by CIMMYT. The objective of the LAMA aflatoxin trials was to identify exotic LAMA lines with reduced susceptibility to aflatoxin accumulation that are suitable for use in a maize breeding program, to compare agronomic characteristics of the LAMA testcrosses to US hybrids to evaluate the usefulness of the LAMA lines in a breeding program, and to compare the response of the LAMA lines when combined with different testers to begin determining how these exotic lines fit into US maize heterotic groups. The objective of the CIMMYT aflatoxin trials is to identify maize inbred lines and hybrids that accumulate low levels of aflatoxin in individual

environments in the Southern US and lines that exhibit tolerance across locations. The objective of the grain composition experiment was to compare the content of exotic adapted hybrids, advanced yellow testcrosses, and quality protein maize hybrids to US hybrid maize.

CHAPTER II

LAMA AFLATOXIN TRIALS

Introduction

The US maize germplasm base is rather narrow, a characteristic typical of agricultural systems in developed countries (Smith, 1988). The lack of genetic diversity in US maize means that genes for certain specific traits like resistance to specific insects and diseases are lacking in US germplasm. Tropical germplasm has often been cited as a source of new genes for disease and insect resistance as well as other desirable agronomic traits (Geadelmann, 1984; Albrecht et al., 1987). Tropical germplasm may be a good source of resistance to aflatoxin since no US germplasm has adequate genetic resistance.

Aflatoxin, produced by *Aspergillus flavus* fungus, is a potent toxin and carcinogen that can cause aflatoxicosis and liver cancer in humans and animals (Payne, 1992). Aflatoxin affects crops worldwide especially in tropical regions (Park and Liang, 1993). Approximately 25% of the world's food supply is contaminated by aflatoxin according to an FAO estimate (Moreno and Kang, 1999). In the US, aflatoxin is a chronic problem in the south while only a periodic problem in the Corn Belt (Payne, 1992; Betran et al., 2002).

Aflatoxin limits the marketability of contaminated maize grain because of the risk to human and animal health and corresponding regulation (Betran et al., 2002). The estimated economic loss reaches hundreds of millions of dollars in years of severe outbreaks (Moreno and Kang, 1999).

Genetic variation exists in maize for response to *A. flavus* indicating that resistance to aflatoxin accumulation is heritable (Payne, 1992; Betran and Isakeit, 2004). Genetic resistance may be the most effective control strategy for *Aspergillus* ear and kernel rot (Campbell and White, 1995a). Adequate genetic resistance is not available in any current commercial hybrids. No control strategy has been effective in years when environmental conditions are extremely favorable for *A. flavus* development (Payne, 1992).

More than 50 countries have established or proposed regulations controlling aflatoxin levels in food and feed (Cleveland et al., 2003). Many countries established a tolerance of 10 ng g^{-1} aflatoxin in food supplies while other countries adopted a zero tolerance policy for aflatoxin contamination (Windham et al., 1999). In the United States, grain with aflatoxin levels exceeding 20 ng g^{-1} is banned from interstate commerce and grain with more than 300 ng g^{-1} aflatoxin cannot be fed to livestock (USDA, 1994).

Aflatoxin contamination has been associated with biotic and abiotic stresses (Gorman et al., 1992; Payne, 1992; Betran et al., 2002). Aflatoxin development is favored by drought conditions and above average temperatures (McMillian et al., 1985; Payne, 1992; Betran et al., 2002). Soil fertility and weed related stresses (Munkvold, 2003) and insect damage to developing ears (McMillian et al., 1985; Windham et al., 1999) have been associated with increased aflatoxin development. Environmental conditions like drought appear more influential than insect damage (Munkvold, 2003).

Earliness is not sufficient to reduce aflatoxin accumulation. Overall adaptation of a hybrid to its environment appears to be more important in minimizing aflatoxin contamination. Adaptation indicated by increased grain yield is correlated to reduced aflatoxin content in hybrids (Betran and Isakeit, 2004).

No control methods currently available adequately control aflatoxin accumulation in years with optimal conditions for development of *A. flavus.* Fungicides are ineffective at controlling aflatoxin (Brown et al., 1999). Biocontrol using atoxigenic strains of *A. flavus* to competitively exclude aflatoxin producing strains may help reduce aflatoxin contamination in regions with chronic problems (Cleveland et al., 2003). Irrigation to avoid drought stress was found to affect airborne inoculum loads, kernel infection, aflatoxin content at harvest, and yields (Jones, 1987). Subsoiling to reduce drought stress was also found to result in less aflatoxin contaminated grain (Payne, 1992).

In general, cultural practices should be managed to reduce as much plant stress as possible, especially during silking. Factors to control include field selection, planting date, crop rotation, tillage, plant density, soil fertility, and irrigation (Jones, 1987; Payne, 1992; Munkvold, 2003). Balanced soil fertility programs (Jones, 1987) and increased nitrogen fertilization (Munkvold, 2003) were found to minimize aflatoxin contamination of maize. Planting locally adapted maize hybrids reduces the risk of abiotic stress and aflatoxin accumulation (Payne, 1992; Munkvold, 2003).

The exact nature of aflatoxin resistance is unknown, but some resistant genotypes have a thick waxy covering on the kernels or differences in kernel proteins when

compared to susceptible lines (Guo et al., 1995; Windham and Williams, 2002; Munkvold, 2003), good husk coverage (Payne, 1992; Betran et al., 2002; Munkvold, 2003), resistance to insect damage (Payne, 1992; Betran et al., 2002), and ears that do not remain upright (Munkvold, 2003) were found to correlate with reduced aflatoxin accumulation.

Several well-characterized sources of aflatoxin resistance exist (Munkvold, 2003), but high levels of resistance have not been incorporated into commercially valuable hybrids (Campbell and White, 1995a; Betran and Isakeit, 2004). The majority of aflatoxin resistance sources lack agronomic performance (Betran and Isakeit, 2004). Much of the resistant germplasm identified matures too late for use in the Corn Belt (Campbell and White, 1995a, 1995b; Munkvold, 2003). Only a limited amount of germplasm has been evaluated for resistance to aflatoxin (Gorman et al., 1992; Campbell and White, 1995b).

Progress in developing aflatoxin resistant genotypes has also been hampered by the fact that no specific resistance factors have been identified (Guo et al., 1995) and there is a lack of single major gene resistance (Munkvold, 2003). While aflatoxin production is influenced by the genetics of the plant, it appears to be quantitatively inherited with significant additive and non-additive effects (Gardner et al., 1987). Other challenges facing breeders include:

1. Disease development is greatly influenced by the environment (Darrah et al., 1987; Payne, 1992; Campbell and White, 1995b).

2. Natural outbreaks of *A. flavus* are sporadic so natural infection is unreliable for resistance screening (Campbell and White, 1995a).
3. Inconsistent and labor intensive inoculation techniques (Payne, 1992; Campbell and White, 1995b; Munkvold, 2003).
4. Lack of genotypes to use as resistant control (Munkvold, 2003).
5. Expense of aflatoxin assays, no way to visually evaluate lines for aflatoxin resistance (Payne, 1992; Munkvold, 2003).
6. Results are sensitive to sampling procedure (Campbell and White, 1995b).
7. Resistance must be expressed in a mature plant in a low metabolic state thus limiting the types of resistance to preformed compounds and morphological barriers (Payne, 1992).

Tropical maize tends to be poorly adapted to the US and pose significant challenges to inclusion in a US maize breeding program. One strategy to overcome these challenges is to develop tropical lines adapted to temperate environments by selecting for earliness, reduced lodging, and short plant heights. Using this method the resulting lines are 100% tropical lines that are adapted to temperate growing conditions.

Using this strategy of adaptation, early generation maize lines were obtained from commercial maize companies in South America. Selections were made for earliness, reduced plant and ear height, reduced lodging, and lower grain moisture. Selections were made in Weslaco, a semi-tropical environment, and College Station, a semi-temperate environment. Selections were successful in reducing root and stalk lodging, grain moisture, and plant height (Ochs, 2005). The resulting lines, known as

LAMA lines, were testcrossed to LH195 and LH210, elite temperate testers developed by Holden's Foundation Seeds. LH195 is a stiff stalk type tester and LH210 is a non-stiff stalk plant type.

The primary objective of this study is to identify exotic LAMA lines with reduced susceptibility to aflatoxin accumulation that are suitable for use in a maize breeding program. Other research objectives include comparing agronomic characteristics of the LAMA lines to US hybrids to evaluate the usefulness of the LAMA lines in a breeding program, and to compare the response of the LAMA lines when combined with different testers, a stiff-stalk type and a non-stiff-stalk type, to begin determining how these exotic lines fit into US maize heterotic groups.

Materials and Methods

The LAMA testcrosses were grown in field experiments in multiple Texas environments including College Station, Weslaco, Corpus Christi, Bardwell, Wharton, and Dalhart in 2005 and 2006. In 2005 14 different LAMA lines testcrossed to LH195 and LH210 and 4 US hybrids were included in the trial of 30 total entries. In 2006, 60 total entries included 39 LAMA lines testcrossed to LH195 and LH210 and 6 US hybrids. The entries in the 2006 trial were selections from the 2005 trial and a 2005 yield trial that was not evaluated for aflatoxin accumulation. All field trials were planted using a replicated alpha-lattice experimental design (Patterson and Williams, 1976). All plots were two row plots planted to a population of 70,000 plants per hectare in irrigated environments and 60,000 plants per hectare in dryland environments. Agronomic traits

including plant and ear height, grain yield, grain moisture, and test weight were measured in all locations. Aflatoxin accumulation was evaluated in College Station, Weslaco, and Corpus Christi. In College Station and Weslaco, 10 randomly selected plants per plot were inoculated using the non-wounding silk channel inoculation method. In Corpus Christi, plots were inoculated using the colonized kernel method (Odvody et al., 2000). In addition, this location was planted a month later than normal to induce stress on the plants and encourage growth of *A. flavus*. Inoculated ears were hand harvested and the aflatoxin was quantified using the Vicam system (Betran et al., 2002). All data collected was analyzed using Remltool for single location analysis and SAS proc GLM for across location analysis. Data from single locations was analyzed using ANOVA and least significant differences were used to differentiate the performance of genotypes. The data was combined across locations and analyzed using ANOVA and orthogonal contrasts were run to identify differences between the LAMA testcrosses and US hybrids. The stability of LAMA testcrosses across environments was determined using the lsd and rank shifts between genotypes.

The environments at College Station, Corpus Christi, and Weslaco differ in rainfall and temperature representing the maize growing conditions in Texas (Table 2.1). In 2006 Corpus Christi had more rain than in 2005, but the field trial received 21.74 cm of rain in one day accounting for almost half of the rainfall for the growing season. College Station receives more rain than the other locations in typical years. The length of the growing season varied from 135 to 160 days so growing degree days (GDD) accumulated per day is reported to show the difference in air temperature between

environments. Corpus Christi accumulated more GDD/day than the other environments when calculated with an upper limit of 30 degrees Celsius and when calculated with no upper limit. Weslaco accumulated slightly more GDD/day than College Station, but those locations were more similar than Corpus Christi.

Table 2.1. Weather data for trial locations

Location	**College Station**		**Weslaco**		**Corpus Christi**	
Year	**2005**	**2006**	**2005**	**2006**	**2005**	**2006**
Rainfall (cm)	33.71	33.25	10.67	8.84	18.44	46.48[c]
Irrigation (cm)	22.86	15.24	50.80	50.80	0.00	0.00
Total Moisture (cm)	56.57	48.49	61.47	59.64	18.44	46.48
GDD (ºC)	6530.72	6351.89	5827.67	5601.22	5848.33	6032.53
Days in Field	160	155	142	135	135	138
GDD/Day[a]	23.17	23.33	23.39	23.83	25.67	26.07
GDD/Day[b]	24.34	24.29	24.37	24.47	27.11	27.35

[a] GDD calculated with upper limit of 30 degrees C and a lower limit of 10 degrees C

[b] GDD calculated with no upper limit

[c] 21.74 cm rainfall on 6-1-06

Results and Discussion

The LAMA lines had lower grain yields, higher grain moisture, lower aflatoxin accumulation, and higher test weights than the US hybrids (Table 2.2). The LAMA lines were not significantly different than the US hybrids for plant height, ear height, or lodging.

Table 2.2. Orthogonal contrasts of LAMA testcrosses versus US hybrids for log transformation of aflatoxin accumulation, yield, moisture, plant height, ear height, lodging, and test weight

	2005			2006		
Trait	df	Mean Square	Pr>F	df	Mean Square	Pr>F
Log aflatoxin	1	1.55133	0.0004	1	7.06361	<.0001
Yield	1	142.29501	<.0001	1	59.07305	<.0001
Moisture	1	421.26738	<.0001			
Plant height	1	1.85387	0.8954			
Ear height	1	0.15881	0.9647			
Lodging	1	49.35991	0.3744			
Test weight	1	9.59630	<.0001			

Across all environments, the LAMA testcrosses had consistently lower aflatoxin accumulation than the US hybrids (Figure 2.1). This observation suggests that the LAMA lines have aflatoxin resistance factors not present in current US maize hybrids.

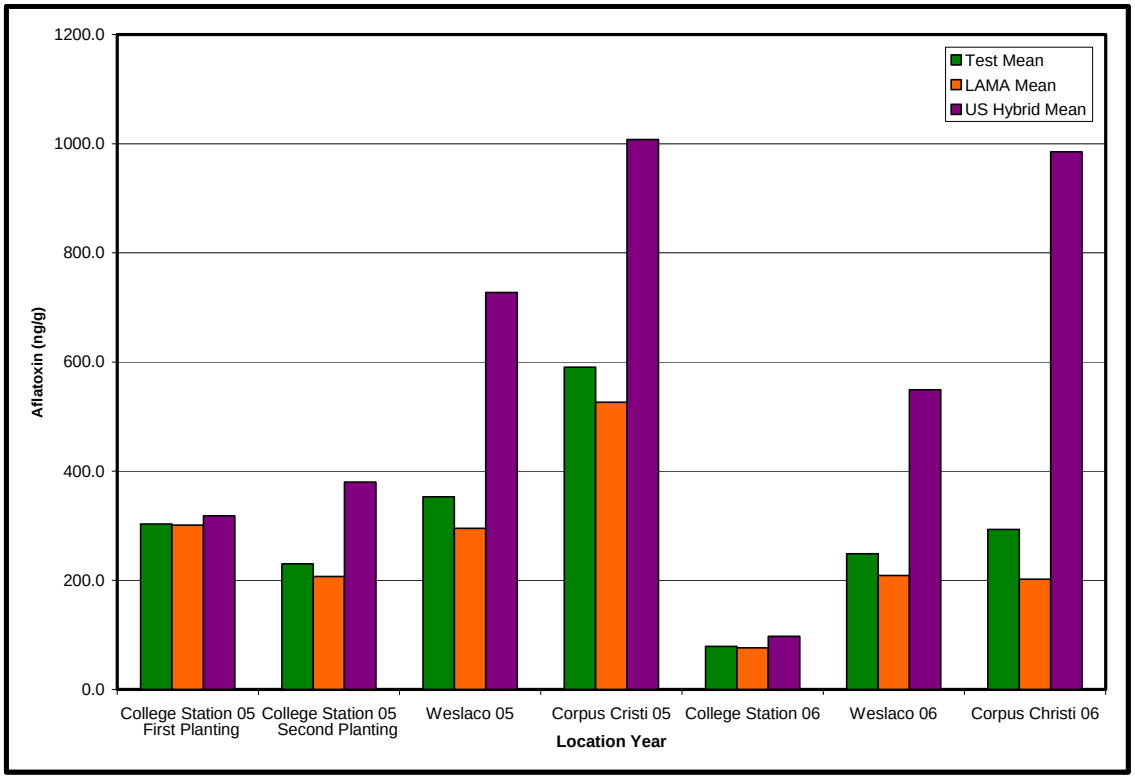

Figure 2.1. Mean Aflatoxin Accumulation for all Hybrids, LAMA Testcrosses, and US Hybrids across Locations and Years

All LAMA testcrosses were developed with two elite temperate testers, Holdens lines LH195 and LH210. The LAMA testcrosses with LH195 accumulated less aflatoxin than the LAMA testcrosses with tester LH210 (Figure 2.2). An orthogonal contrast shows this difference in testers was significant in 2005 (df=1, mean square=2.61776, Pr>F=<.0001), but non-significant in 2006 (df=1, mean square=0.10225, Pr>F=0.4237).

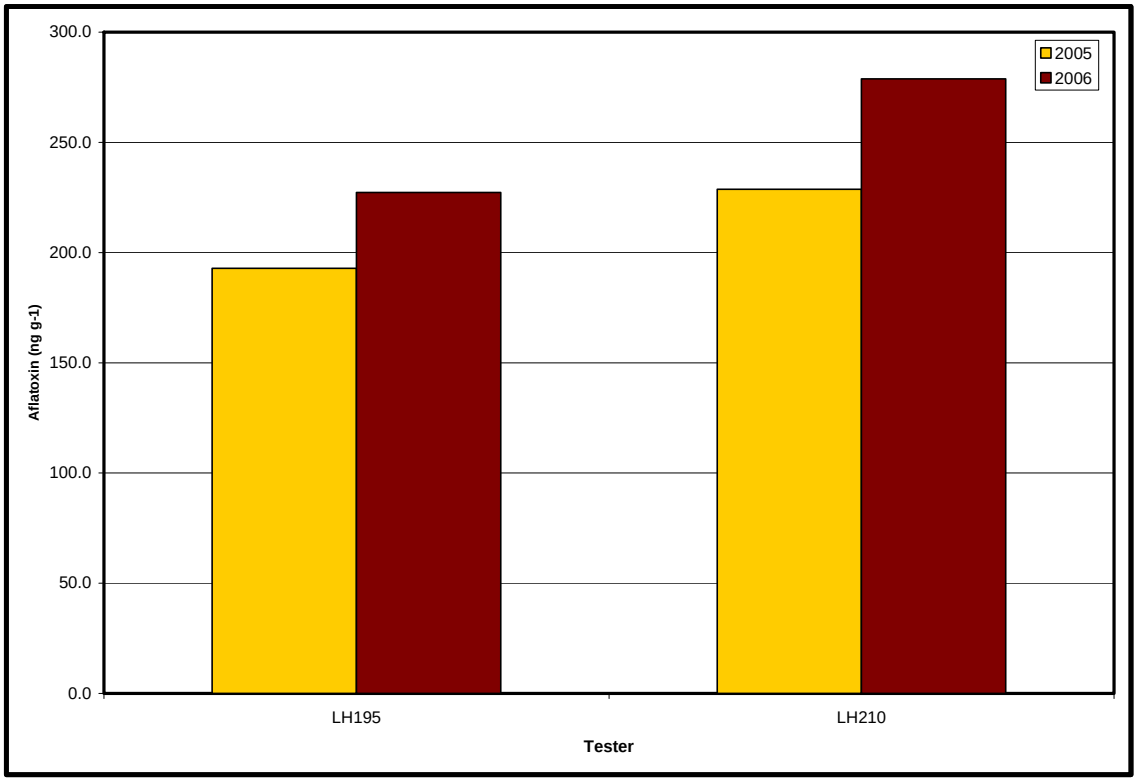

Figure 2.2. Mean Aflatoxin Accumulation for LAMA Testcrosses by Tester in 2005 and 2006 Field Trials

Significant differences for aflatoxin accumulation were observed in the 2005 trials (Table 2.3). Across locations, the top performing LAMA testcrosses were entries L5/LH195 (112.7 ng g^{-1} aflatoxin), L7/LH195 (140.5 ng g^{-1}), and L3/LH195 (175.6 ng g^{-1}). US hybrid P32R25 (1287.2 ng g^{-1}) accumulated the most aflatoxin across locations, followed by LAMA testcross entry L2/LH210 (786.3 ng g^{-1}), US hybrids DKC 69-72 (673.0 ng g^{-1}) and P31B13 (562.0 ng g^{-1}), and LAMA testcross entries L8/LH210 (557.2 ng g^{-1}) and L10/LH210 (535.1 ng g^{-1}). The mean aflatoxin accumulation across locations was 391.18 ng g^{-1}. DKC 69-70 was the only US hybrid to accumulate less

aflatoxin than the test mean in each location and across locations. Entry L5/LH195 was in the top three ranking testcrosses in the Weslaco and Corpus Christi locations.

LAMA testcross entries L12/LH195 (72.4 ng g^{-1}), L8/LH195 (75.9 ng g^{-1}), and L8/LH210 (95.5 ng g^{-1}) were the least susceptible entries to aflatoxin accumulation in the trial at College Station where the test mean was 230.25 ng g^{-1}. Eighteen LAMA testcrosses and one US hybrid (DKC69-70) were not significantly different than entry L12/LH195. US hybrid P31B13 accumulated the most aflatoxin in College Station (528.8 ng g^{-1}), while entry L13/LH195 (429.8 ng g^{-1}) was the most susceptible LAMA testcross.

In Weslaco, 15 LAMA testcrosses and one US hybrid (DKC 69-70) were not significantly different than entry L7/LH195 (91.9 ng g^{-1}), the most aflatoxin resistant entry in the trial. P32R25 accumulated the most aflatoxin (1412.5 ng g^{-1}).

The Corpus Christi trial had the highest mean (590.37 ng g^{-1}). Eight LAMA testcrosses and US hybrid DKC 69-70 were not significantly different than the least susceptible hybrid L5/LH195 (133.3 ng g^{-1}). US hybrid P32R25 (2060.2 ng g^{-1}) accumulated the most aflatoxin in the Corpus Christi trial.

Table 2.3. Aflatoxin accumulation for LAMA testcrosses and US corn hybrids in each location and across locations in 2005

		College Station		Weslaco		Corpus Christi		Across
Entry	Pedigree	AF* Rank	Antilog AF	AF Rank	Antilog AF	AF Rank	Antilog AF	Antilog AF
			ng g^{-1}		ng g^{-1}		ng g^{-1}	ng g^{-1}
L1/LH195	LAMA-1-1-B/LH195	13	167.2a-g	9	210.5a-f	22	616.9d-h	331.5
L2/LH195	LAMA-1-3-B/LH195	25	389.0e-g	4	119.3a-c	12	408.9b-g	305.7
L2/LH210	LAMA-1-3-B-B/LH210	21	297.4b-g	25	528.8e-h	29	1532.9hi	786.3
L3/LH195	LAMA-1-5-B/LH195	15	189.1a-g	6	133.9a-d	4	203.9a-c	175.6
L3/LH210	LAMA-1-5-B-B/LH210	6	110.5a-d	26	533.0e-h	21	608.6d-h	417.3
L4/LH195	LAMA-2-1-B/LH195	20	238.1a-g	5	131.8a-d	2	165.2ab	178.4
L4/LH210	LAMA-2-1-B/LH210	12	158.5a-g	18	352.1b-g	26	940.2g-i	483.6
L5/LH195	LAMA-10-1-B/LH195	5	106.3a-d	2	98.5a	1	133.3a	112.7
L6/LH195	LAMA-11-1-B/LH195	7	117.5a-e	8	168.5a-e	11	385.7b-g	223.9
L6/LH210	LAMA-11-1-B/LH210	19	215.4a-g	23	457.1d-h	17	522.9c-h	398.5
L7/LH195	LAMA-12-1-B/LH195	9	132.8a-f	1	91.9a	3	196.7a-c	140.5
L7/LH210	LAMA-12-1-B/LH210	26	389.0e-g	28	645.7f-h	16	491.7c-g	508.8
L8/LH195	LAMA-25-5-B/LH195	2	75.9a	21	407.4c-h	19	567.2c-h	350.1
L8/LH210	LAMA-25-5-B/LH210	3	95.5ab	27	537.0e-h	28	1039.0g-i	557.2
L9/LH195	LAMA-34-1-B/LH195	14	189.1a-g	12	232.6a-g	15	481.0c-g	300.9
L9/LH210	LAMA-34-1-B/LH210	8	122.1a-e	10	227.4a-g	8	307.1a-e	218.9
L10/LH195	LAMA-42-B-B/LH195	4	104.7a-c	14	259.0a-g	6	264.4a-d	209.4
L10/LH210	LAMA-42-B-B/LH210	24	386.1e-g	13	236.2a-g	27	982.9g-i	535.1
L11/LH195	LAMA-42-B-B/LH195	22	344.1c-g	3	107.2ab	9	325.1a-f	258.8
L11/LH210	LAMA-42-B-B/LH210	18	210.5a-g	16	295.1a-g	7	290.3a-d	265.3
L12/LH195	LAMA-46-3-B/LH195	1	72.4a	11	229.1a-g	5	253.9a-d	185.2
L12/LH210	LAMA-46-3-B/LH210	23	360.3d-g	24	471.3d-h	20	595.0d-h	475.5
L13/LH195	LAMA-58-1-B/LH195	29	429.8fg	20	398.1c-h	18	560.7c-h	462.9

Table 2.3 Continued

Entry	Pedigree	College Station AF Rank	College Station Antilog AF ng g^{-1}	Weslaco AF Rank	Weslaco Antilog AF ng g^{-1}	Corpus Christi AF Rank	Corpus Christi Antilog AF ng g^{-1}	Across Antilog AF ng g^{-1}
L13/LH210	LAMA-58-1-B/LH210	16	190.5a-g	15	273.3a-g	25	908.9f-i	457.6
L14/LH195	LAMA-60-9-B/LH195	10	138.0a-f	7	156.1a-e	13	423.3b-g	239.1
L14/LH210	LAMA-60-9-B/LH210	11	157.3a-g	19	377.3b-g	14	476.0c-g	336.9
P31B13	P31B13	30	528.8g	22	439.8d-h	23	717.3d-i	562.0
P32R25	P32R25	27	389.0e-g	30	1412.5h	30	2060.2i	1287.2
DKC 69-70	DKC 69-70	17	204.2a-g	17	304.3a-g	10	384.5a-g	297.7
DKC 69-72	DKC 69-72	28	398.1e-g	29	752.8gh	24	868.0e-i	673.0

*AF is abbreviation for aflatoxin

In 2005, the College Station field trials had the lowest mean aflatoxin accumulation (230.25 ng g^{-1}) while the Corpus Christi trials had the highest mean aflatoxin accumulation (590.37 ng g^{-1}) (Table 2.4).

Table 2.4. Statistics for 2005 field trials for aflatoxin accumulation trait in each location in Texas

	College Station		Weslaco		Corpus Christi	
	LogAF	Antilog AF	LogAF	Antilog AF	logAF	Antilog AF
Mean	2.30	230.25	2.45	352.92	2.68	590.37
MSE	0.10	.	0.11	.	0.07	.
LSD	0.53	.	0.55	.	0.45	.
Sig.	0.03	.	0.00	.	0.00	.
CV	13.82	.	13.39	.	9.88	.
Min	1.86	72.44	1.96	91.90	2.12	133.26
Max	2.72	528.81	3.15	1412.54	3.31	2060.16

Table 2.5. Analysis of variance across environments for logarithmic transformation of aflatoxin

		LogAflatoxin	
Source	df	Mean Square	Pr>F
Env	2	3.390	<.0001
Rep(Env)	6	0.809	<.0001
Genotype	29	0.341	<.0001
GEI	58	0.106	0.5778

Genotype by environment interaction (GEI) was non-significant in the 2005 field trial (table 2.5). Eight LAMA testcrosses, entries L3/LH195, L4/LH195, L5/LH195, L7/LH195, L9/LH210, L10/LH195, L11/LH210, and L12/LH195 and US hybrid DKC

69-70 were not significantly different than the best performing hybrid in all three environments. Entry L2/LH210 and US hybrids P31B13, P32R25, and DKC 69-72 were not significantly different than the most susceptible hybrid in all three environments. Other entries were not as stable across environments. L13/LH210 was not different than the best hybrid in College Station and Weslaco, but was not different than the worst hybrid in Corpus Christi. Entries L3/LH210 and L8/LH195 were not different than the best hybrid in College Station, but were not different than the worst hybrid in Weslaco. Entry L11/LH195 was not different than the best entry in Weslaco and Corpus Christi, but was not different than the worst hybrid in College Station. Entries L8/LH210, L2/LH195, and L10/LH210 were also not stable across environments.

In 2006, 54 LAMA testcrosses and 6 US hybrids were evaluated for response to aflatoxin. Nineteen LAMA testcrosses accumulated less than 100 ppb aflatoxin across locations (Table 2.6). The top performing testcrosses were entries L28/LH195, L23/LH195, and L30/LH195. The entries accumulating the most aflatoxin across locations were US hybrids P31G66, P31B13, and DKC 66-80. The LAMA testcrosses accumulating the most aflatoxin were entries L15/LH210, L4/LH195, and L45/LH195. The mean aflatoxin accumulation across locations was 148.3 ng g^{-1}. All of the US hybrids accumulated more than the mean for each trial across locations.

In the 2006 College Station trial, two LAMA testcrosses, entries L24/LH210 (293.0 ng g^{-1}) and L4/LH195 (287.7 ng g^{-1}) accumulated more aflatoxin than the most susceptible US hybrid, P31B13 (250.3 ng g^{-1}). Entries L41/LH195 (13.7 ng g^{-1}), L2/LH195 (14.1 ng g^{-1}), and L48/LH195 (20.3 ng g^{-1}) accumulated the least amount of

aflatoxin. US hybrids DKC 66-80 (136.9 ng g^{-1}) and P31G66 (60.2 ng g^{-1}) were not significantly different than the best performing entry in the trial.

In the Weslaco trial, US hybrid P31B13 (1176.8 ng g^{-1}) accumulated the most aflatoxin in the trial, significantly more than any other entry. LAMA testcross entries L41/LH210 (852.5 ng g^{-1}), L15/LH210 (648.3 ng g^{-1}), and L24/LH210 (635.9 ng g^{-1}) were the LAMA lines accumulating the most aflatoxin in Weslaco while entries L32/LH195 (28.0 ng g^{-1}), L40/LH195 (37.4 ng g^{-1}), and L17/LH195 (38.4 ng g^{-1}) were the hybrids with the least aflatoxin accumulation.

In the 2006 trial at Corpus Christi, LAMA testcross entry L15/LH210 accumulated 1487.0 ng g^{-1} aflatoxin, more than twice the amount of aflatoxin accumulated by entry L44/LH210 (643.0 ng g^{-1}), the LAMA testcross accumulating the next highest amount of aflatoxin. US hybrid W4700 (276.3 ng g^{-1}) was the only US hybrid to accumulate less than the mean aflatoxin accumulation of 293.3 ng g^{-1} for the test. LAMA testcross entries L23/LH195 (30.2 ng g^{-1}), L28/LH195 (34.3 ng g^{-1}), and L21/LH210 (47.0 ng g^{-1}) were the three top performing entries for aflatoxin.

Table 2.6. Aflatoxin accumulation for LAMA testcrosses and US hybrids in each location in 2006

		College Station		Weslaco		Corpus Christi	
Entry	Pedigree	AF rank	Antilog AF	AF rank	Antilog AF	AF rank	Antilog AF
			ng g^{-1}		ng g^{-1}		ng g^{-1}
L2/LH195	LAMA-1-3-B/LH195	2	14.1ab	12	71.8a-g	28	179.6c-m
L4/LH195	LAMA-2-1-B/LH210	59	287.7g	54	567.3lm-o	52	448.3k-r
L8/LH195	LAMA-25-5-B/LH195	28	58.3a-g	6	45.8a-c	7	77.9a-f
L14/LH210	LAMA-60-9-B/LH210	13	34.3a-f	51	431.1j-o	34	223.0d-o
L15/LH210	Tx732-B-B-B/LH210	10	28.3a-e	58	648.3m-o	59	1487.0qr
L16/LH195	LAMA-1-2-B/LH195	8	28.0a-e	10	65.1a-f	24	152.9b-l
L17/LH195	LAMA-2-2-B/LH195	44	91.1d-g	3	38.4ab	27	170.1c-m
L18/LH210	LAMA-2-3-B/LH210	53	134.3fg	50	430.7j-o	32	205.9c-n
L19/LH195	LAMA-2-6-B/LH195	35	70.3c-g	18	113.4b-j	36	231.9d-p
L19/LH210	LAMA-2-6-B/LH210	32	61.9b-g	36	215.0e-m	30	182.4c-m
L20/LH195	LAMA-2-7-B/LH195	23	49.5a-g	30	191.2d-m	47	331.7f-p
L20/LH210	LAMA-2-7-B/LH210	39	79.7c-g	23	140.7b-k	20	112.3a-k
L21/LH195	LAMA-5-3-B/LH195	7	27.7a-e	15	94.1a-h	10	88.8a-h
L21/LH210	LAMA-5-3-B/LH210	52	119.3e-g	37	215.4e-m	3	47.0a-c
L22/LH210	LAMA-5-5-B/LH210	31	61.9b-g	56	618.2m-o	5	64.2a-d
L23/LH195	LAMA-7-2-B/LH195	20	43.0a-g	11	66.1a-f	1	30.2a
L24/LH210	LAMA-9-2-B/LH210	60	293.0g	57	635.9m-o	39	255.0d-p
L25/LH195	LAMA-9-3-B/LH195	57	213.3g	41	240.3e-n	41	272.1d-p
L25/LH210	LAMA-9-3-B/LH210	48	108.4e-g	31	201.4d-m	33	220.9d-o
L26/LH195	LAMA-14-B-B/LH195	15	35.0a-f	40	224.0e-n	14	99.3a-j
L26/LH210	LAMA-14-B-B/LH210	22	49.1a-g	21	130.9b-k	4	48.6a-c
L27/LH195	LAMA-17-3-B/LH195	11	29.8a-e	29	184.9d-m	49	369.5h-r
L27/LH210	LAMA-17-3-B/LH210	12	34.2a-f	42	249.4f-n	40	264.7d-p
L28/LH195	LAMA-20-3-B/LH195	33	64.0c-g	9	62.7a-e	2	34.3ab

Table 2.6. Continued

		College Station		Weslaco		Corpus Christi	
Entry	Pedigree	AF rank	Antilog AF	AF rank	Antilog AF	AF rank	Antilog AF
			ng g^{-1}		ng g^{-1}		ng g^{-1}
L29/LH195	LAMA-20-4-B/LH195	4	22.8a-d	4	45.1a-c	23	144.5b-k
L29/LH210	LAMA-20-4-B/LH210	43	84.6c-g	17	111.5b-j	13	99.2a-j
L30/LH195	LAMA-20-5-B/LH195	6	27.1a-e	35	213.9e-m	8	83.2a-g
L31/LH210	LAMA-22-2-B/LH210	19	38.2a-f	49	420.8i-o	25	153.4c-m
L32/LH195	LAMA-22-3-B/LH195	42	84.2c-g	1	28.0a	22	133.3a-k
L32/LH210	LAMA-22-3-B/LH210	55	172.2g	45	303.9h-o	11	91.5a-i
L33/LH195	LAMA-22-5-B/LH195	18	37.0a-f	24	153.5c-l	26	165.3c-m
L33/LH210	LAMA-22-5-B/LH210	50	117.1e-g	14	94.0a-h	19	112.2a-k
L34/LH195	LAMA-23-3-B/LH195	27	57.1a-g	26	168.0c-m	48	351.4g-q
L35/LH195	LAMA-34-7-B/LH195	24	51.3a-g	7	53.2a-d	29	180.8c-m
L36/LH195	LAMA-35-2-B-B/LH195	37	73.5c-g	16	106.3a-i	46	329.8f-p
L37/LH195	LAMA-35-4-B/LH195	47	99.3d-g	34	208.6d-m	44	317.4f-p
L37/LH210	LAMA-35-4-B/LH210	40	79.9c-g	47	340.8h-o	6	67.8a-e
L38/LH210	LAMA-35-5-B/LH210	45	92.1d-g	38	218.5e-n	45	327.9f-p
L39/LH210	LAMA-40-B-B/LH210	26	57.0a-g	39	220.4e-n	17	110.7a-k
L40/LH195	LAMA-44-B-B/LH195	9	28.0a-e	2	37.4ab	18	111.4a-k
L40/LH210	LAMA-44-B-B/LH210	54	143.8fg	32	203.3d-m	15	105.6a-k
L41/LH195	LAMA-46-6-B/LH195	1	13.7a	19	116.4b-k	51	439.9j-r
L41/LH210	LAMA-46-6-B/LH210	56	190.2g	59	852.5no	16	108.7a-k
L42/LH195	LAMA-53-1-B/LH195	41	83.0c-g	22	139.4b-k	12	97.4a-i
L43/LH195	LAMA-53-4-B/LH195	29	59.5a-g	48	402.1i-o	21	123.3a-k
L44/LH195	LAMA-53-5-B/LH195	36	71.3c-g	43	263.1g-n	50	393.8i-r
L44/LH210	LAMA-53-5-B/LH210	25	54.4a-g	44	269.3g-n	54	643.0l-r
L45/LH195	LAMA-56-B-B/LH195	5	23.4a-d	33	207.5d-m	53	621.3l-r
L45/LH210	LAMA-56-B-B/LH210	16	36.4a-f	28	181.5d-m	37	235.3d-p

Table 2.6. Continued

		College Station		Weslaco		Corpus Christi	
Entry	**Pedigree**	**AF rank**	**Antilog AF**	**AF rank**	**Antilog AF**	**AF rank**	**Antilog AF**
			ng g^{-1}		**ng g^{-1}**		**ng g^{-1}**
L46/LH195	LAMA-58-2-B/LH195	21	43.9a-g	5	45.2a-c	43	298.0e-p
L47/LH195	LAMA-58-5-B/LH195	46	94.9d-g	13	89.3a-h	31	188.1c-m
L47/LH210	LAMA-58-5-B/LH210	34	69.1c-g	20	125.8b-k	38	243.4d-p
L48/LH195	LAMA-58-7-B/LH195	3	20.3a-c	8	61.8a-e	9	84.1a-g
L49/LH195	LAMA-61-2-B/LH195	14	34.7a-f	25	166.5c-m	35	226.8d-p
B-H 8913	B-H 8913	51	117.1e-g	46	337.3h-o	55	686.0m-r
DKC66-80	DKC66-80	17	36.9a-f	53	459.2k-o	60	1635.3r
DKC69-71	DKC69-71	49	108.6e-g	27	179.6d-m	56	882.3n-r
P31B13	P31B13	58	250.3g	60	1176.8o	57	912.9o-r
P31G66	P31G66	30	60.2a-g	55	606.7l-o	58	1017.2p-r
W4700	W4700	38	78.7c-g	52	435.3j-o	42	276.3d-p

There were significant differences in aflatoxin accumulation in each location and across locations (table 2.7). The College Station test had the lowest mean (78.8 ng g^{-1}), minimum (13.7 ng g^{-1}), and maximum (293.0 ng g^{-1}) aflatoxin accumulation while Corpus Christi had the highest mean (293.3 ng g^{-1}), minimum (30.2 ng g^{-1}), and maximum (1635.3 ng g^{-1}).

Table 2.7. Statistics for 2006 field trials for aflatoxin accumulation in each location and across locations for corn hybrids

	College Station		Weslaco		Corpus Christi		Across	
	Log AF*	Antilog AF ng g^{-1}	Log AF	Antilog AF ng g^{-1}	Log AF	Antilog AF ng g^{-1}	Log AF	Antilog AF ng g^{-1}
Mean	1.79	78.8	2.25	248.8	2.29	293.3	2.11	148.2
MSE	0.15	.	0.13	.	0.14	.	0.48	.
LSD	0.65	.	0.59	.	0.64	.	.	.
Sig	0.01	.	0.00	.	0.00	.	0.02	.
CV	21.63	.	15.74	.	16.43	.	18.99	.
Min	1.14	13.7	1.45	28.0	1.48	30.2	1.61	41.2
Max	2.47	293.0	3.07	1176.8	3.21	1635.3	2.61	411.6

*AF=Aflatoxin

Table 2.8. Analysis of variance across environments for log aflatoxin data transformation

Source	df	LogAflatoxin	
		Mean Square	Pr>F
Env	2	14.018	<.0001
Rep(Env)	6	0.112	0.6521
Genotype	59	0.475	0.0202
GEI	118	0.304	<.0001

Genotype by environment interaction (GEI) was significant in the 2006 field trials (table 2.8). LAMA testcross entries L8/LH195, L21/LH195, L23/LH195, L40/LH195, and L48/LH195 were not significantly different than the least susceptible line in all three locations. LAMA testcross L4/LH195 and US hybrids B-H 8913 and P31B13 were not significantly different than the worst line in all three locations.

Other LAMA testcrosses were not stable across environments. Entries L14/LH210, L15/LH210, L27/LH195, L31/LH210, L41/LH195, L43/LH195, L44/LH210, L45/LH195 and US hybrids DKC 66-80 and P31G66 were not significantly different than the line accumulating the least aflatoxin in College Station, but were not significantly different than the entry accumulating the most aflatoxin in one of the other locations. Testcrosses L17/LH195, L28/LH195, L32/LH195, L33/LH210, L36/LH195, and L47/LH195 were not significantly different than the best hybrid in Weslaco, but were not significantly different than the worst hybrid in College Station. The final group of entries that were not stable across environments did not accumulate significantly more aflatoxin than the least susceptible line in Corpus Christi, but were not significantly different than the most susceptible line in College Station or Weslaco. These entries included L20/LH210, L21/LH210, L22/LH210, L28/LH195, L29/LH210, L32/LH195, L32/LH210, L33/LH210, L37/LH210, L40/LH210, L41/LH210, and L42/LH195.

As a group, the exotic LAMA lines consistently yield less grain than the US hybrids across locations and years, but some individual testcrosses are competitive with US hybrids for yield (Figure 2.3). The Corpus Christi 2005 field trial had the lowest

mean yield, that trial was planted late and not irrigated to induce plant stress and encourage aflatoxin colonization.

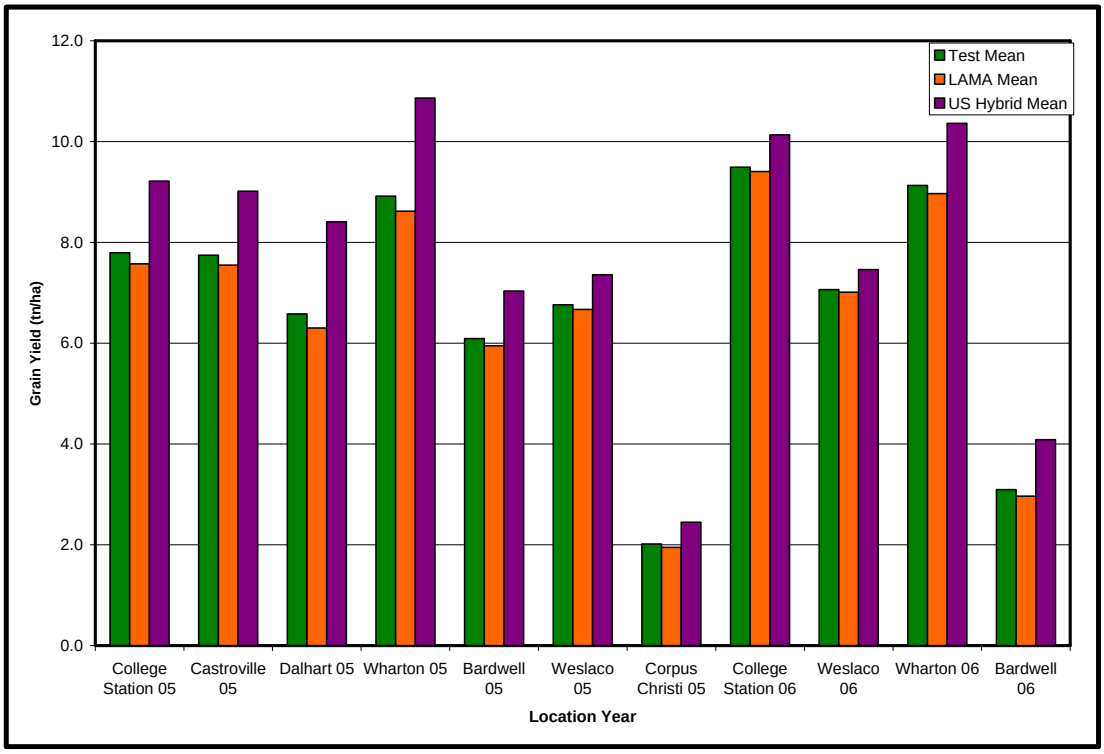

Figure 2.3. Mean Grain Yield for Test, LAMA Testcrosses, and US Hybrids Across Locations and Years

Orthogonal contrasts show that there is no significant difference in yield between testcrosses with tester LH195 and tester LH210 (2005 df=1, mean square=0.01801, Pr>F=.8599; 2006 df=1, mean square=1.19780, Pr>F=.2827).

Table 2.9. Grain yield in all locations and across locations in 2005 for commercial US hybrids and hybrids between LAMA lines and LH195 and LH210

Entry	College Station Mg ha^{-1}	Weslaco Mg ha^{-1}	Corpus Christi Mg ha^{-1}	Castroville Mg ha^{-1}	Dalhart Mg ha^{-1}	Wharton Mg ha^{-1}	Bardwell Mg ha^{-1}	Across Mg ha^{-1}
L1/LH195	7.0	6.2	1.5	7.6	6.8	8.8	5.6	6.22
L2/LH195	6.5	6.4	2.0	7.4	7.3	9.3	4.5	6.19
L2/LH210	7.8	6.2	1.7	5.4	3.7	9.1	5.2	5.58
L3/LH195	6.6	7.3	2.1	7.9	7.7	6.9	5.4	6.27
L3/LH210	6.4	5.7	2.3	7.1	4.1	9.8	6.2	5.94
L4/LH195	6.7	7.4	2.6	7.8	5.8	8.6	5.1	6.30
L4/LH210	6.1	5.6	1.9	6.9	5.6	7.7	5.4	5.61
L5/LH195	7.1	7.1	1.8	6.9	6.8	9.0	6.9	6.52
L6/LH195	7.1	6.1	1.5	6.3	6.1	7.5	5.3	5.70
L6/LH210	7.4	5.8	2.1	7.6	7.5	8.5	6.3	6.46
L7/LH195	8.2	6.9	1.7	7.9	5.6	9.8	6.5	6.68
L7/LH210	7.5	6.6	2.1	8.3	6.8	8.0	6.9	6.61
L8/LH195	8.6	7.3	1.9	8.1	6.8	8.8	5.9	6.79
L8/LH210	7.4	6.6	2.2	7.7	6.0	8.4	5.7	6.29
L9/LH195	7.4	6.4	1.5	7.3	6.7	8.0	5.7	6.15
L9/LH210	7.8	6.4	1.5	7.9	6.3	7.1	5.5	6.09
L10/LH195	8.1	7.2	1.5	7.5	6.9	9.0	5.6	6.55
L10/LH210	7.3	7.3	2.2	8.1	4.5	9.9	6.7	6.58
L11/LH195	8.4	7.0	1.8	7.0	6.2	8.7	6.5	6.49
L11/LH210	8.7	7.2	1.8	8.5	8.3	9.3	7.4	7.32
L12/LH195	7.9	6.5	1.9	7.7	6.2	8.3	4.6	6.14
L12/LH210	7.9	5.8	2.5	8.1	4.7	8.2	5.9	6.16
L13/LH195	7.9	7.3	1.6	7.8	7.2	9.0	6.9	6.83
L13/LH210	9.0	7.0	2.4	8.0	5.5	8.7	7.0	6.81
L14/LH195	8.2	7.1	2.0	7.6	8.5	8.9	5.8	6.85
L14/LH210	8.1	7.1	2.4	7.7	6.2	8.7	5.9	6.60

Table 2.9. Continued

Entry	College Station Mg ha^{-1}	Weslaco Mg ha^{-1}	Corpus Christi Mg ha^{-1}	Castroville Mg ha^{-1}	Dalhart Mg ha^{-1}	Wharton Mg ha^{-1}	Bardwell Mg ha^{-1}	Across Mg ha^{-1}
DKC 69-72	10.2	7.8	2.7	9.2	7.0	11.8	7.1	7.97
P31B13	9.3	7.3	2.8	9.3	8.7	10.6	7.0	7.86
P32R25	8.6	6.3	2.0	8.6	8.7	10.2	7.6	7.42
W4700	.	.	.	8.9	9.2	10.9	6.5	7.81
Mean	7.79	6.76	2.01	7.75	6.58	8.92	6.09	6.55
MSE	0.63	0.19	0.10	0.29	1.07	0.48	0.35	.
LSD	1.44	0.76	0.57	0.97	1.84	1.22	1.01	.
Sig.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	.
CV	0.00	6.37	15.80	6.95	15.73	7.73	9.74	.
Min	6.14	5.56	1.46	5.38	3.75	6.86	4.50	5.58
Max	10.23	8.00	2.83	9.35	9.18	11.76	7.58	7.97

Entry L11/LH210 was the only LAMA testcross to average yield greater than 7 Mg ha-1 across locations (Table 2.9).

In College Station, LAMA testcross entries L45/LH195 (12.5 Mg ha^{-1}) and L2/LH195 (12.3 Mg ha^{-1}) yielded significantly more grain than US hybrids DKC66-80 (9.1 Mg ha^{-1}) and W4700 (8.7 Mg ha^{-1}) and not significantly different than the other US hybrids in 2006 (Table 2.10). US hybrid DKC69-71 yielded significantly more than all other entries in the trials at Weslaco and Wharton. L36/LH195 (8.4 Mg ha^{-1}), the highest yielding LAMA testcross in Weslaco, yielded significantly more than the US hybrids DKC66-80 (7.1 Mg ha^{-1}), P31B13 (7.3 Mg ha^{-1}), P31G66 (6.8 Mg ha^{-1}), and W4700 (7.3 Mg ha^{-1}). In the Wharton field trial, the grain yield of LAMA testcross entry L35/LH195 (10.3 Mg ha^{-1}) was not significantly different than any of the US hybrids except for DKC69-71 (11.8 Mg ha^{-1}). Entry L27/LH210 (4.5 Mg ha^{-1}) yielded significantly more than US hybrid DKC69-71 and not significantly different than the other US hybrids in the Bardwell trials.

Table 2.10. Grain yield across locations 2006 for commercial US hybrids and hybrids between LAMA lines and LH195 and LH210

Entry	Pedigree	College Station		Weslaco		Wharton		Bardwell		Across locations
		Yield rank	Grain yield Mg ha^{-1}	Yield Rank	Grain yield Mg ha^{-1}	Yield rank	Grain yield Mg ha^{-1}	Yield rank	Grain yield Mg ha^{-1}	Grain yield Mg ha^{-1}
L2/LH195	LAMA-1-3-B/LH195	2	12.3	11	7.8	9	10.1	45	2.6	8.3
L4/LH195	LAMA-2-1-B/LH210	32	9.4	59	5.4	56	7.8	11	3.8	6.5
L8/LH195	LAMA-25-5-B/LH195	40	9.1	19	7.5	21	9.6	19	3.4	7.6
L14/LH210	LAMA-60-9-B/LH210	12	10.6	22	7.5	44	8.7	51	2.5	7.2
L15/LH210	Tx732-B-B-B/LH210	52	8.2	52	6.0	42	8.8	4	4.2	7
L16/LH195	LAMA-1-2-B/LH195	39	9.1	42	6.7	53	8.0	36	3.0	6.6
L17/LH195	LAMA-2-2-B/LH195	23	9.8	18	7.6	18	9.7	35	3.0	7.7
L18/LH210	LAMA-2-3-B/LH210	60	6.9	60	4.9	60	6.9	17	3.5	5.5
L19/LH195	LAMA-2-6-B/LH195	18	10.3	27	7.4	16	9.9	23	3.3	7.6
L19/LH210	LAMA-2-6-B/LH210	37	9.1	53	6.0	48	8.4	26	3.2	6.6
L20/LH195	LAMA-2-7-B/LH195	5	11.1	26	7.4	28	9.3	24	3.3	7.9
L20/LH210	LAMA-2-7-B/LH210	42	9.0	56	5.7	35	9.0	27	3.2	6.6
L21/LH195	LAMA-5-3-B/LH195	20	10.0	16	7.6	46	8.6	25	3.3	7.5
L21/LH210	LAMA-5-3-B/LH210	9	10.8	46	6.4	41	8.8	29	3.2	7.3
L22/LH210	LAMA-5-5-B/LH210	30	9.5	43	6.6	47	8.5	43	2.7	6.7
L23/LH195	LAMA-7-2-B/LH195	21	9.9	5	7.9	12	10.0	42	2.7	7.7
L24/LH210	LAMA-9-2-B/LH210	43	8.9	39	6.8	51	8.2	22	3.4	6.8
L25/LH195	LAMA-9-3-B/LH195	27	9.6	10	7.8	38	8.9	38	2.8	7.4
L25/LH210	LAMA-9-3-B/LH210	15	10.4	41	6.7	29	9.2	30	3.1	7.3
L26/LH195	LAMA-14-B-B/LH195	36	9.2	15	7.7	22	9.5	56	2.2	7.1
L26/LH210	LAMA-14-B-B/LH210	58	7.4	50	6.2	39	8.9	12	3.7	6.7
L27/LH195	LAMA-17-3-B/LH195	26	9.6	38	7.0	15	9.9	9	3.9	7.7

Table 2.10. Continued

		College Station		Weslaco		Wharton		Bardwell		Across
Entry	Pedigree	Yield rank	Grain yield Mg ha^{-1}	Yield Rank	Grain yield Mg ha^{-1}	Yield rank	Grain yield Mg ha^{-1}	Yield rank	Grain yield Mg ha^{-1}	Grain yield Mg ha^{-1}
L27/LH210	LAMA-17-3-B/LH210	28	9.5	54	5.9	50	8.3	2	4.5	7.0
L28/LH195	LAMA-20-3-B/LH195	54	7.9	25	7.4	43	8.7	57	2.1	6.5
L29/LH195	LAMA-20-4-B/LH195	41	9.0	14	7.7	45	8.6	40	2.8	7.1
L29/LH210	LAMA-20-4-B/LH210	45	8.7	47	6.3	52	8.1	14	3.6	6.6
L30/LH195	LAMA-20-5-B/LH195	35	9.4	4	8.0	19	9.6	59	1.7	7.3
L31/LH210	LAMA-22-2-B/LH210	57	7.5	51	6.1	25	9.4	34	3.0	6.5
L32/LH195	LAMA-22-3-B/LH195	47	8.6	28	7.4	40	8.9	54	2.3	6.7
L32/LH210	LAMA-22-3-B/LH210	50	8.3	37	7.1	55	7.8	15	3.6	6.6
L33/LH195	LAMA-22-5-B/LH195	16	10.3	21	7.5	23	9.5	52	2.4	7.4
L33/LH210	LAMA-22-5-B/LH210	51	8.2	58	5.5	49	8.4	20	3.4	6.5
L34/LH195	LAMA-23-3-B/LH195	48	8.5	55	5.8	54	8.0	21	3.4	6.3
L35/LH195	LAMA-34-7-B/LH195	7	10.9	13	7.8	5	10.3	47	2.7	7.7
L36/LH195	LAMA-35-2-B-B/LH195	19	10.1	2	8.4	32	9.1	37	2.9	7.5
L37/LH195	LAMA-35-4-B/LH195	53	8.0	7	7.9	27	9.3	53	2.4	6.8
L37/LH210	LAMA-35-4-B/LH210	25	9.6	12	7.8	31	9.1	16	3.5	7.6
L38/LH210	LAMA-35-5-B/LH210	59	6.9	48	6.2	59	7.2	39	2.9	5.7
L39/LH210	LAMA-40-B-B/LH210	14	10.4	34	7.1	20	9.6	31	3.1	7.5
L40/LH195	LAMA-44-B-B/LH195	33	9.5	44	6.5	36	8.9	33	3.1	6.8
L40/LH210	LAMA-44-B-B/LH210	56	7.5	49	6.2	57	7.5	8	4.0	6.2
L41/LH195	LAMA-46-6-B/LH195	22	9.9	31	7.2	14	9.9	60	1.3	7.1
L41/LH210	LAMA-46-6-B/LH210	31	9.5	36	7.0	30	9.2	48	2.6	7.1
L42/LH195	LAMA-53-1-B/LH195	8	10.9	6	7.9	34	9.0	50	2.6	7.7

Table 2.10. Continued

		College Station		Weslaco		Wharton		Bardwell		Across
Entry	Pedigree	Yield rank	Grain yield Mg ha^{-1}	Yield Rank	Grain yield Mg ha^{-1}	Yield rank	Grain yield Mg ha^{-1}	Yield rank	Grain yield Mg ha^{-1}	Grain yield Mg ha^{-1}
L43/LH195	LAMA-53-4-B/LH195	17	10.3	17	7.6	10	10.0	55	2.2	7.5
L44/LH195	LAMA-53-5-B/LH195	29	9.5	24	7.4	37	8.9	28	3.2	7.2
L44/LH210	LAMA-53-5-B/LH210	34	9.4	57	5.6	58	7.4	13	3.6	6.7
L45/LH195	LAMA-56-B-B/LH195	1	12.5	23	7.5	8	10.2	49	2.6	8.2
L45/LH210	LAMA-56-B-B/LH210	49	8.3	45	6.4	17	9.9	10	3.8	7.1
L46/LH195	LAMA-58-2-B/LH195	24	9.7	35	7.1	33	9.0	41	2.8	7.3
L47/LH195	LAMA-58-5-B/LH195	44	8.8	20	7.5	24	9.5	46	2.7	7.1
L47/LH210	LAMA-58-5-B/LH210	55	7.8	32	7.2	26	9.4	32	3.1	6.9
L48/LH195	LAMA-58-7-B/LH195	10	10.8	9	7.9	13	10.0	58	2.0	7.6
L49/LH195	LAMA-61-2-B/LH195	13	10.6	8	7.9	11	10.0	44	2.7	7.9
B-H 8913	B-H 8913	4	11.5	3	8.2	3	10.6	7	4.0	8.4
DKC69-71	DKC69-71	3	11.7	1	9.4	1	11.8	18	3.4	9.0
DKC66-80	DKC66-80	38	9.1	33	7.1	6	10.2	3	4.4	7.7
P31B13	P31B13	6	11.1	29	7.3	2	10.6	6	4.0	8.2
P31G66	P31G66	11	10.7	40	6.8	7	10.2	5	4.1	8.0
W4700	W4700	46	8.7	30	7.3	4	10.6	1	4.5	7.6
Mean			9.492		7.063		9.132		3.097	7.198
MSE			1.951		0.194		0.300		0.256	.
LSD			2.407		0.748		1.097		1.012	.
Sig			0.000		0.000		0.000		0.000	.
CV			14.715		6.231		6.002		16.329	.
Min			6.913		4.874		6.900		1.250	5.500
Max			12.531		9.412		11.750		4.500	9.000

The LAMA lines are later maturing lines than the US hybrids included in the field trials and consistently have higher grain moisture than the US hybrids in all locations and years (figure 2.4).

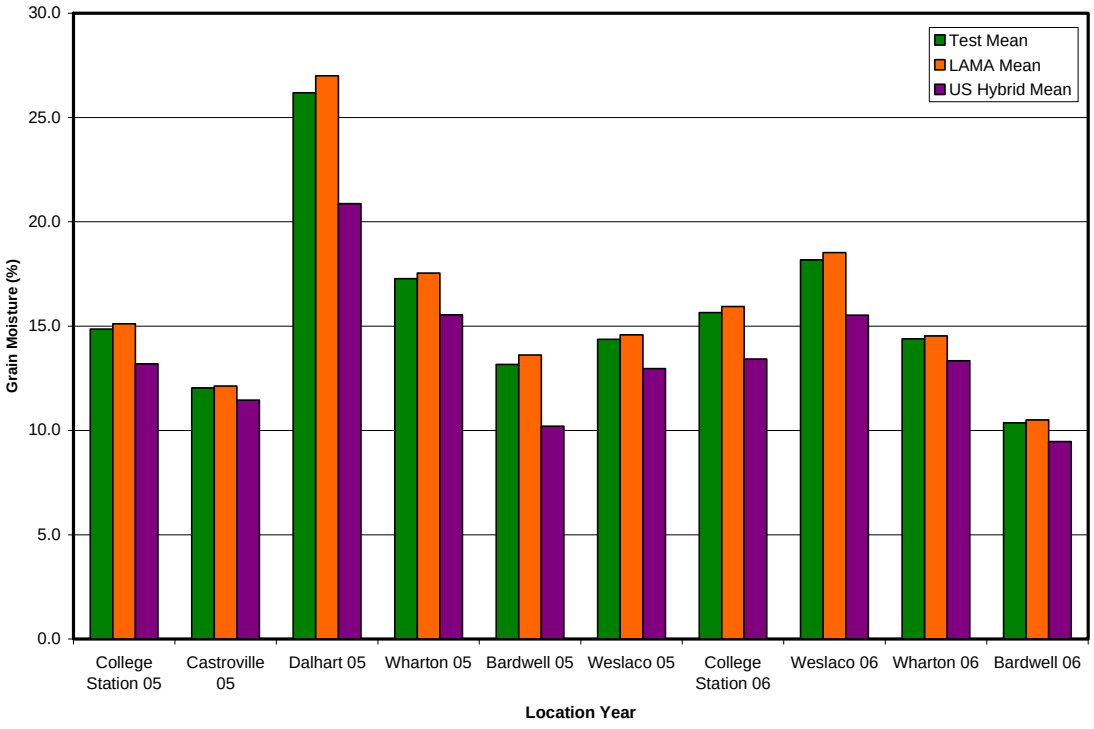

Figure 2.4. Mean Grain Moisture for the Test, LAMA Testcrosses, and US Hybrids across Locations and Years.

Analysis of variance for agronomic traits (table 2.11 and table 2.12) showed that environment is significant for all traits, lodging, ear height, plant height, moisture, yield, and test weight. GEI is significant for all traits except plant height. Genotype was significant for grain moisture, grain yield, and test weight, but not for lodging, ear height, and plant height.

Table 2.11. ANOVA for Agronomic Traits in 2005 field trials.

	Lodging			Ear height			Plant height	
Source	df	Mean Square	Pr>F	df	Mean Square	Pr>F	df	Mean Square	Pr>F
Env	3	6904.45956	<.0001	1	31162.84090	<.0001	1	26480.83593	<.0001
Rep(Env)	8	170.83421	0.0066	3	397.59777	0.0033	3	152.73566	0.2388
Genotype	29	165.81141	0.3155	29	237.37958	0.0910	29	295.44966	0.0575
GEI	87	145.66710	<.0001	29	143.62226	0.0212	29	163.02427	0.0680

	Moisture			Yield			Test Weight	
Source	df	Mean Square	Pr>F	df	Mean Square	Pr>F	df	Mean Square	Pr>F
Env	5	2378.01479	<.0001	6	441.08070	<.0001	5	166.19556	<.0001
Rep(Env)	12	5.36548	<.0001	14	3.16636	<.0001	12	0.86388	0.0431
Genotype	29	27.19949	<.0001	29	7.85285	<.0001	29	6.30430	0.0005
GEI	145	5.32598	<.0001	174	1.45645	<.0001	145	2.67810	<.0001

Table 2.12. ANOVA for Agronomic Traits in 2006 trials.

	Moisture			Yield			Test Weight	
Source	df	Mean Square	Pr>F	df	Mean Square	Pr>F	df	Mean Square	Pr>F
Env	3	1152.99384	<.0001	3	1123.43622	<.0001	3	313.16756	<.0001
Rep(Env)	6	1.59517	0.0806	6	3.80974	0.0015	6	1.46219	0.4137
Genotype	59	10.12541	<.0001	59	4.03162	<.0001	59	7.56585	<.0001
GEI	170	1.31789	0.0003	177	1.79803	<.0001	170	2.16033	0.0009

Conclusions

The mean aflatoxin accumulation of the exotic adapted LAMA lines was consistently lower than the mean accumulation of the US hybrids across locations and years. This trend suggests that the LAMA lines have aflatoxin resistance factors not present in current US hybrids.

Across years, the College Station trial had the lowest mean and lowest maximum aflatoxin accumulation while Corpus Christi consistently had the highest aflatoxin accumulation. In 2005, the mean aflatoxin accumulation in College Station was 230.25 ng g^{-1} while the trial mean in Corpus Christi was 590.37, more than twice the mean of the College Station trial. In 2006 the trial mean in Corpus Christi, 293.3 ng g^{-1}, was over three and a half times as great as the College Station mean of 78.8ng g^{-1}.

In the 2006 field trials, genotype by environment interaction was significant and several LAMA testcrosses appear better adapted to specific environments within the region while other testcrosses were stable across environments. LAMA testcross entries L8/LH195, L21/LH195, L23/LH195, L40/LH195, and L48/LH195 were not significantly different than the best line in all three locations. Few LAMA testcrosses were included in both years of field trials. These testcrosses need to be evaluated across more years to study stability within an environment across years.

The mean grain yield for the LAMA testcrosses was lower than the mean yield of the US hybrids in all locations and years of the field trials. Large differences were observed between environments. In 2005, Corpus Christi had the lowest trial mean of 2.01 Mg ha^{-1} while Wharton had the highest trial mean of 8.92 Mg ha^{-1}. In 2006, the

lowest trial mean of 3.1 Mg ha^{-1} was observed in Bardwell while the highest mean of 9.5 Mg ha^{-1} was produced in College Station. Individual LAMA testcrosses are competitive with US hybrids and have potential to contribute to a breeding program.

The LAMA testcrosses have higher mean grain moisture and test weight than the US hybrids. Selection during line development was effective for lowering grain moisture although the LAMA lines still have higher grain moisture than the US hybrids. Selection during line development for resistance to lodging has been effective at reducing the percentage of lodged plants in the LAMA material.

CHAPTER III

CIMMYT AFLATOXIN TRIALS

Introduction

Aflatoxin is a chronic problem across the Southern US and around the world in locations with hot, dry climates. To combat this problem, CIMMYT (International Center for Maize and Wheat Improvement) has developed advanced inbreds and hybrids with reduced susceptibility to aflatoxin that adapt to these regions. The objectives of this study were to identify maize inbred lines and hybrids developed by CIMMYT in Mexico that accumulate low levels of aflatoxin in individual environments in the Southern US and lines that exhibit tolerance to aflatoxin across locations.

Materials and Methods

This study is a collaborative project between CIMMYT, where the inbred lines and hybrids were selected for low aflatoxin accumulation, and maize programs at Texas A&M University and USDA in Georgia and Mississippi, the three programs that evaluated the inbred lines and hybrids in the Southern US. CIMMYT developed the 70 inbred lines, 23 yellow hybrids, and 30 white hybrids and sent seed for the field trials that were evaluated in three locations across the Southern US: College Station, Texas; Tifton, Georgia; and Starkville, Mississippi. Local US inbreds and hybrids were included in the trials at each location. The material evaluated was divided into three

aflatoxin trials in each location: inbred lines, yellow hybrids, and white hybrids. A description of the US inbreds and hybrids is included in table A.1 in the appendix and a description of the CML lines included in the inbred and hybrid trials is included in table A.2. The CML lines were developed from lowland tropical germplasm and subtropical germplasm and are intermediate to late maturing. Some of the CML lines are QPM (quality protein maize) lines and CML 341, CML 342, CML 343, and CML 344 are drought tolerant. All of the CIMMYT lines were selected in Mexico for reduced susceptibility to *A. flavus* colonization and low aflatoxin accumulation.

All entries were grown in an alpha lattice experimental design with two reps per location. All field trials were irrigated as needed and managed for maximum grain yield. The field plots were inoculated with *Aspergillus flavus* isolate NRRL 3357. Inoculation method varied by location. The tests in Texas were inoculated using the non-wounding silk channel inoculation technique (Scott and Zummo, 1988). The tests in Mississippi were inoculated using the side needle method (Scott and Zummo, 1988), and the Georgia tests were inoculated using the knife method (Widstrom et al., 1981).

Aflatoxin was quantified using Vicam Aflatest columns. Single location analysis was conducted with Remltool and across location analysis was completed using SAS Proc mixed. ANOVA was performed on single location data and combined data. Least significant difference was calculated to identify differences between genotypes in individual locations.

Results and Discussion

Inbred line – Combined Analysis

Genotype, environment, replication within environment, and genotype by environment interaction were all significant across locations (Table 3.1). Entries CML 451 and LPSC3-54-1-2-2-3-B-B-B-B-B-B -B-B accumulated high amounts of aflatoxin in the Georgia trial, but appear to be better adapted to the environment in Mississippi where LPSC3-54-1-2-2-3-B-B-B-B-B-B -B-B was the second ranking entry in the test. Lines CL-04353, CL-02844, CML 341, and CL-02510 performed better in the Georgia test. Entries Mp313E, La Posta Seq C7-F180-1-2-2-1-B-B, CL-RCW37, and CL-RCW35 responded similarly to both environments, but tended to not be among the best performing lines in either location.

Table 3.1. ANOVA for the inbred trial across locations in 2006.

		LogAflatoxin	
Source	df	Mean Square	Pr>F
Env	1	2.01150788	<.0001
Rep(Env)	5	0.25479652	0.0330
Genotype	69	0.62892542	0.0033
GEI	56	0.30833664	<.0001

Inbred Line Tests by Location

In the Tifton, GA trial, the mean aflatoxin accumulation was 921.586 ng g^{-1}, and significant differences were observed among the entries (Table 3.2). The entries CL-04353 (64.88 ng g^{-1}), CML 161 (65.28 ng g^{-1}), and CL-SCBY03 (81.87 ng g^{-1})

accumulated the least aflatoxin in Georgia. Fourteen entries did not accumulate significantly more aflatoxin than CL-04353, the top performing entry in the test. The entries CML 159 (8448.9 ng g^{-1}), CML 451 (6985.54 ng g^{-1}), and P390Bco/CML c4 F24-B-1-2-1-B-B (5245.66 ng g^{-1}) accumulated the most aflatoxin in the Georgia inbred trials.

In Starkville, MS, aflatoxin accumulation averaged 1287.89 ng g^{-1} across all entries (Table 3.2). The entries CML 176 (81.41 ng g^{-1}), LPSC3-54-1-2-2-3-B-B-B-B-B-B-B-B (147.03 ng g^{-1}), and CML 495, CL-RCW01 (202.58 ng g^{-1}) accumulated the least aflatoxin in the Mississippi inbred trial. Only four lines did not accumulate significantly more aflatoxin than CML 176, the top performing line in the trial. The most susceptible entries in the test include SC 212M (20849.71 ng g^{-1}) which accumulated significantly more aflatoxin than any other entry in the test, La Posta Seq C7-F64-1-1-1-2-B-B (3682.14 ng g^{-1}), and La Posta Seq C7-F64-1-1-1-1-B-B, (3575.20 ng g^{-1}).

Table 3.2. Aflatoxin accumulation of inbreds in Georgia and Mississippi in 2006.

	Georgia		Mississippi	
Pedigree	Log AF	Antilog AF	Log AF	Antilog AF
CML 247	2.18	150.42a-i	2.38	239.33b-f
CML 342	.	.	2.69	494.31c-m
DTPWC8F31-1-3-1-B-B-B-B	2.82	663.74k-w	2.56	365.09b-i
DTPWC8F266-1-1-1-B-B	3.04	1093.7n-w	2.46	289.93b-g
SPLC7F254-1-2-3-2-2-B-B-B-B	.	.	3.23	1681.90p-t
P43C9-56-1-1-1-2-B-B-B-B-B	3.17	1489.02q-x	3.16	1431.53n-t
CML 254	.	.	.	.
CML 341	1.98	95.68a-d	2.81	652.53f-p
CML 343	3.18	1525.81r-x	3.24	1750.25p-t
G16BNSEQC0F118-1-1-4-2-B-B-B-B-B	2.91	814.33m-w	2.46	287.67b-g
LPSC3-54-1-2-2-3-B-B-B-B-B-B -B-B	3.24	1722.66u-x	2.17	147.03ab
CML 448	2.49	312.54b-o	3.08	1190.42l-r
CL-RCW35	3.20	1577.97s-x	3.18	1528.97o-t
CML 449	2.62	413.24e-s	2.58	382.03b-j
CML 495, CL-RCW01	2.56	363.66d-p	2.31	202.58a-c
CML 494, CL-04365	2.61	403.27e-r	3.27	1878.02q-t
CLFAWW11	2.35	222.89a-m	2.68	474.57c-m
CML 451	3.84	6985.54yz	2.64	432.91c-l
CL-02844	2.66	461.32e-u	3.52	3322.00st
CL-02603	1.95	88.57a-c	.	.
CL-02450	2.44	278.55b-m	2.74	548.15c-n
CML 481	2.47	295.26b-n	2.57	369.15b-i
CML 269	2.72	527.11f-v	2.96	905.94h-r
CML 384	3.21	1614.73t-x	2.76	577.17e-o
CL-04353	1.81	64.88a	2.70	502.00c-m
CL-04343	2.74	555.78h-v	2.32	210.43a-e
CML 492	2.53	339.78c-p	3.01	1031.57j-r
La Posta Seq C7-F86-3-1-1-1-B-B	2.55	356.37d-p	.	.
La Posta Seq C7-F103-2-2-2-2-B	2.93	858.22m-w	3.16	1447.10n-t

Table 3.2. Continued

	Georgia		Mississippi	
Pedigree	Log AF	Antilog AF	Log AF	Antilog AF
CLQ-RCWQ80	.	.	.	.
CLQ-RCWQ80	2.08	120.09a-e	.	.
CML 150	2.50	313.18b-o	2.56	360.74b-i
CML 159	3.93	8448.9z	3.25	1767.67p-t
CLQ-RCWQ82	2.68	481.95f-u	2.74	549.54c-n
[(P390bcoC3 F191-1-1-1-4-B-B-B-B) x (P73TLC3#-115-1-4-#)]-1-2-2-B-B-B	2.53	338.38c-p	2.76	569.64d-o
La Posta Seq C7-F64-1-1-1-1-B-B	3.40	2503.23w-z	3.55	3575.20t
La Posta Seq C7-F64-1-1-1-2-B-B	3.28	1905.02v-y	3.57	3682.14t
P390bcoC3/254/247 F29-1-2-1-B-B-B	2.62	412.76e-s	2.75	562.21d-o
La Posta Seq C7-F180-1-2-2-1-B-B	3.23	1685.00u-x	3.24	1726.63p-t
La Posta Seq C7-F96-1-1-1-1-B-B	3.20	1596.98s-x	3.10	1256.90m-s
P390Bco/CML c4 F24-B-1-2-1-B-B	3.72	5245.66x-z	.	.
La Posta Seq C7-F179-3-4-1-1-B-B	2.62	418.22e-t	2.74	549.41c-n
CL-SPLW04	2.24	171.87a-k	2.74	548.91c-n
CL-04368	3.11	1279.38p-w	2.56	363.75b-i
CL-RCW37	2.88	750.93l-w	2.86	730.30g-q
CL-02510	2.42	262.85b-m	3.22	1674.94p-t
CML493, CLQ-6601	2.14	138.80a-f	2.74	543.25c-n
CLQ-G2507	2.22	164.36a-i	2.35	223.05b-e
CLQ-6311	2.75	566.37i-v	3.10	1251.41m-s
CML 454	2.14	139.28a-g	2.48	300.26b-g
CML 479	3.07	1161.45o-w	3.24	1743.81p-t
CML 144	2.65	448.23e-u	2.97	924.49i-r
CML 450	2.48	302.27b-o	2.75	559.24d-o
CML 161	1.81	65.28a	2.52	332.89b-h
CML 163	2.72	527.23f-v	.	.
CML 172	.	.	3.15	1405.72n-t
MIRTC5 Bco F24-2-1-2-1-1-B-B	2.58	382.3e-q	2.60	397.47b-k
CML 344	.	.	.	.

Table 3.2. Continued

	Georgia		Mississippi	
Pedigree	Log AF	Antilog AF	Log AF	Antilog AF
P21STEC1HC27-1-6-1-1-4-BBB-2-##-B*9-B-B	2.76	570.43i-v	.	.
P25C5HC246-3-1-BB-2-#-B*11-B-B	.	.	3.04	1092.70k-r
CML 176	2.46	287.21b-n	1.91	80.41a
P73TLC3# -153-1-1-#-#-B-B-B-B	2.82	666.81k-w	3.38	2376.84r-t
La Posta Seq C7-F31-2-3-1-1-B-B	2.30	197.88a-l	2.42	262.85b-f
La Posta Seq C7-F71-1-1-1-2-B-B	.	.	3.37	2330.24r-t
NC312	3.04	1107.39n-w	.	.
CML285	2.73	541.88g-v	.	.
CML480	2.22	166.72a-j	.	.
CL-RCY016	2.90	789.95m-w	.	.
CL-SCBY03	1.91	81.87ab	.	.
Tzi8	3.22	1678.03u-x	.	.
Tzi18	2.81	645.06j-w	.	.
Mp313E	2.52	329.38c-p	2.52	328.40b-g
T173	2.88	765.42l-w	.	.
Mp717	2.16	143.05a-h	2.73	531.99c-n
Va35	2.83	670.19k-w	.	.
LH195	.	.	3.30	2001.70q-t
SC 212M	.	.	4.32	20849.71u
Mp 04:87	.	.	2.75	556.42c-o
Mp 04:96	.	.	2.32	207.97a-d
Mean	**2.70**	**921.59**	**2.86**	**1287.89**
MSE	**0.12**	**.**	**0.06**	**.**
LSD	**0.59**	**.**	**0.44**	**.**
Sig	**0.00**	**.**	**0.00**	**.**
Min	**1.81**	**64.88**	**1.91**	**80.41**
Max	**3.93**	**8448.90**	**4.32**	**20849.71**

Yellow Hybrid Tests by Location

The Texas trial (51.30 ng g^{-1}) had the lowest mean aflatoxin accumulation of the three environments (Table 3.3). The entries CML451 x CL-07905 (0.98 ng g^{-1}), CML451 x CL-02844 (2.16 ng g^{-1}), and CML287 x CML451 (5.4 ng g^{-1}) accumulated the least aflatoxin in the test. Six entries did not accumulate more aflatoxin than CML451 x CL-07905, the top performing line in the test, and seven entries accumulated less than 20 ng g^{-1} aflatoxin, the limit allowable for interstate commerce of maize grain in the US. Trial entries CLQ-RCYQ44 x CLQ-RCYQ40 (116.98 ng g^{-1}) and (CML451xCL-02450) x CL-RCY015 (128.00 ng g^{-1}) and US hybrid DKC69-71 (160.77 ng g^{-1}) accumulated the most aflatoxin in the test.

The Georgia trial had a mean aflatoxin accumulation of 90.66 ng g^{-1}. Entries CML451 x CL-02844 (31.93 ng g^{-1}), CML287 x CML451 (36.01 ng g^{-1}), and CML480 x CML451 (36.11 ng g^{-1}) had the lowest aflatoxin accumulations in the test. Ten hybrids did not accumulate significantly more aflatoxin than CML451 x CL-02844. Test entries CML451xCL-02450 (155.1 ng g^{-1}) and CML452 x CML451 (201.47 ng g^{-1}) and US hybrid DKC697 (246.72 ng g^{-1}) accumulated the most aflatoxin in the Georgia trial.

The Mississippi trial had the highest location mean aflatoxin accumulation (222.06 ng g^{-1}). The top three performing hybrids, CLQ-RCYQ14 x CML161 (5.41 ng g^{-1}), CML454 x CL-SCBY03 (20.63 ng g^{-1}), and CML451 x CL-07905 (21.53 ng g^{-1}) did not accumulate significantly different amounts of aflatoxin. Entries CML479 x CL-SCBY03 (505.94 ng g^{-1}), CLQ-RCYQ44 x CLQ-RCYQ40 (651.93 ng g^{-1}), and CML453 x CL-SCBY03 (1001.38 ng g^{-1}) accumulated the most aflatoxin in the trial.

Table 3.3. Aflatoxin accumulation for yellow hybrids in each location in 2006.

	Texas		Georgia		Mississippi	
Pedigree	Log AF	Antilog AF	log AF	Antilog AF	Log AF	Antilog AF
CML 492 x CML 491 (Af Resistant Check)	0.7680	5.86a-c	2.0180	104.23c-g	2.1392	137.78c-g
CML451xCML454	1.8898	77.59c-f	1.7917	61.90a-e	2.5668	368.81f-h
CML480 x CML451	1.5749	37.58b-f	1.5576	36.11ab	2.2963	197.83d-h
CML451xCML481	1.9180	82.79c-f	1.8480	70.47a-f	2.4006	251.54e-h
CML451 x CL-02844	0.3344	2.16ab	1.5042	31.93a	1.8174	65.67b-e
CML451xCL-02450	0.9252	8.42a-e	2.1906	155.1f-h	1.9650	92.26b-f
CL-02450 x CML454	1.6101	40.75c-f	1.9553	90.22c-g	1.5362	34.37bc
CML453 x CL-SCBY03	1.8467	70.26c-f	2.1799	151.32f-h	3.0006	1001.38h
CML454 x CL-SCBY03	1.5524	35.68b-f	1.8944	78.42b-f	1.3146	20.63ab
CML479 x CL-SCBY03	1.8769	75.32c-f	1.9456	88.23c-g	2.7041	505.94gh
(CML451xCL-02450) x CML481	1.4902	30.92b-f	1.8587	72.23a-f	1.8521	71.14b-e
(CML451xCL-02450) x CL-RCY015	2.1072	128.00ef	1.8769	75.32b-f	1.5728	37.39bc
(CL-7907xCL-02450) x CML451	2.0330	107.89d-f	2.1120	129.42e-h	2.4834	304.37e-h
CLQ-RCYQ44 x CLQ-RCYQ40	2.0681	116.98ef	1.7205	52.54a-d	2.8142	651.93gh
CML287 x CML451	0.7320	5.40a-c	1.5564	36.01ab	2.4048	253.98e-h
CL-RCY029 x CML451	1.4123	25.84b-f	1.9109	81.45b-f	1.9317	85.45b-f
CML452 x CML451	1.4698	29.5b-f	2.3042	201.47gh	2.2028	159.51c-g
CML451 x CL-07905	0.0071	0.98a	1.5668	36.88ab	1.3330	21.53ab
CLQ-RCYQ14 x CML161	1.0641	11.59a-f	1.6447	44.13a-c	0.7329	5.41a
CLQ-RCYQ19 x CML161	1.7423	55.25c-f	1.9954	98.95c-g	1.9249	84.12b-f
(CML451xCL-02450) x CL-RCY016	2.0666	116.57d-f	2.0630	115.61c-h	2.2318	170.53c-g
(CML451xCML481) x CL-RCY015	0.8143	6.52a-d	1.8991	79.27b-f	1.6374	43.39b-d
(CML451xCML481) x CL-RCY017	1.4059	25.46b-f	1.8069	64.11a-e	2.6154	412.48f-h
P31B13	1.3875	24.41b-f	.	.	.	.
DKC69-71	2.2062	160.77f	.	.	.	.
Mp717 x Mp313E	.	.	1.8096	64.51a-e	.	.

Table 3.3. Continued

	Texas		Georgia		Mississippi	
Pedigree	Log AF	Antilog AF	log AF	Antilog AF	Log AF	Antilog AF
DKC697	.	.	2.3922	246.72h	.	.
Mp 715 x Mp 717	.	.	.	.	1.9256	84.26b-f
GA 209 x SC 212M	.	.	.	.	2.6901	489.89gh
Mean	**1.45**	**51.30**	**1.90**	**90.66**	**2.08**	**222.06**
MSE	**0.35**	**.**	**0.06**	**.**	**0.18**	**.**
LSD	**1.26**	**.**	**0.37**	**.**	**0.71**	**.**
Sig	**0.07**	**.**	**0.00**	**.**	**0.00**	**.**
Min	**-0.01**	**0.98**	**1.50**	**31.93**	**0.73**	**5.41**
Max	**2.21**	**160.77**	**2.39**	**246.72**	**3.00**	**1001.38**

Yellow Hybrid – Combined Analysis

Genotype, environment, replication within environment, and genotype by environment interaction were all significant across locations (table 3.4).

Table 3.4. ANOVA for yellow hybrids across locations in 2006.

		LogAflatoxin	
Source	df	Mean Square	Pr>F
Env	2	5.96358867	<.0001
Rep(Env)	6	0.47258025	0.0064
Genotype	24	1.10176948	0.0015
GEI	48	0.40383008	<.0001

Environment and genotype by environment interaction were both significant indicating entries responded differently to different environments. Hybrid entries CLQ-RCYQ19 x CML161, (CML451xCL-02450) x CL-RCY016, and CL-02450 x CML454 were rather stable across environments. Hybrids CML451 x CL-02844 and CML451 x CL-07905 were the top two performing lines in both the Texas and Georgia environments. CML 492 x CML 491 was one of the top four lines in each location.

White Hybrid – Combined Analysis

Analysis of variance shows that genotype, environment, and genotype by environment interaction are significant for the white hybrid trial while replication within environment was not significant (table 3.5).

Table 3.5. ANOVA for white hybrids across locations in 2006.

		LogAflatoxin	
Source	df	Mean Square	Pr>F
Env	2	22.6799898	<.0001
Rep(Env)	6	0.26260571	0.1408
Genotype	34	1.12154423	0.0228
GEI	66	0.63032286	<.0001

Environment and GEI were significant indicating that hybrids responded differently to the different environments. CIMMYT hybrid CL-RCW35 x CL-04343 was one of the top three lines in Texas and Georgia. Entry CML 495 x CML 343 was one of the top three lines in Texas and Mississippi. The hybrid CML373 x CML384 consistently accumulated large amounts of aflatoxin and was one of the bottom three lines in Texas and Georgia. Genotype x environment was especially important for entry [CML78 x CML373] x TR res EC/MBR IPTT-ECBMo.88-91-2-1-2-(1-6)#b1 16#b 16#b-4-1-B-#-B*11 which was one of the best lines in Mississippi, but one of the three hybrids accumulating the most aflatoxin in Texas.

White Hybrid Tests by Location

This trial included 30 white CIMMYT hybrids and US hybrid checks (Table 3.6). The Texas trial again had the lowest location mean aflatoxin accumulation (28.02 ng g^{-1}). Entries CL-RCW73 x CL-04343 (0.90 ng g^{-1}), CML 495 x CML 343 (1.14 ng g^{-1}), and CL-RCW35 x CL-04343 (1.90 ng g^{-1}) accumulated the least amount of aflatoxin in the test. Seventeen CIMMYT hybrids did not accumulate significantly more aflatoxin than CL-RCW73 x CL-04343 and 18 CIMMYT hybrids accumulated less than

20 ng g^{-1}. CIMMYT hybrids CL-07304 x CML448 (1127.98 ng g^{-1}) , CML373 x CML384 (141.68 ng g^{-1}), and CML264 x CML311] x CML334 (94.67 ng g^{-1}) accumulated the most aflatoxin in the Texas trial.

The Georgia white hybrid trial had a mean aflatoxin accumulation of 156.35 ng g^{-1}). CIMMYT hybrids CL-RCW35 x CL-04343 (31.46 ng g^{-1}), [CML78 x CML373] x TR res EC/MBR IPTT-ECBMo.88-91-2-1-2-(1-6)#b1 16# 16#B1-1-1-B-#-B*6 (35.26 ng g^{-1}), and CL-G2309 x P73TLC3# -153-1-1-#-#-B (41.14 ng g^{-1}) accumulated the least aflatoxin in the trial. Only six hybrids were not significantly different than CL-RCW35 x CL-04343. Entries CML373 x CML384 (558.34 ng g^{-1}), Z1851W (386.63 ng g^{-1}), and Z64W (375.23 ng g^{-1}) accumulated the most aflatoxin in the Georgia test.

The Mississippi trial again had the highest mean aflatoxin accumulation (262.29 ng g^{-1}). The CIMMYT hybrids CML404 x CML476 (7.79 ng g^{-1}), [CML78 x CML373] x TR res EC/MBR IPTT-ECBMo.88-91-2-1-2-(1-6)#b1 16#b 16#b-4-1-B-#-B*11 (19.93 ng g^{-1}), and CML 254 x CML 343 (26.41 ng g^{-1}) accumulated the least aflatoxin in the trial. Six CIMMYT hybrids did not accumulate significantly more aflatoxin than CML404 x CML476 and two of those hybrids accumulated less than 20 ng g^{-1}. Entries (CML264QxCML150) x CML491 (1428.24 ng g^{-1}), CML247 x CML254 (981.75 ng g^{-1}), and the aflatoxin susceptible check CML 491 x CML 150 (900.74 ng g^{-1}) accumulated the most aflatoxin in the Mississippi trial.

Table 3.6. Aflatoxin accumulation for white hybrids in each location in 2006.

	Texas		Georgia		Mississippi	
Pedigree	Log AF	Antilog AF	Log AF	Antilog AF	Log AF	Antilog AF
(CML264 x CML269) x CML449	1.04	10.88a-i	1.98	96.09c-h	1.70	50.52b-d
(P73TLC3# -74-2-6-1-1-#-# x P73TLC3# -241-2-2-1-1-#-#) x CML 448	0.79	6.24a-h	2.02	105.49d-h	2.52	332.35e-j
CL-G2309 x P73TLC3# -153-1-1-#-#-B	0.47	2.98a-e	1.61	41.14a-c	1.88	75.75b-e
CML 449 x CML 448	1.33	21.14b-i	2.06	114.6de-h	1.83	67.98b-e
CML448 x CML449	1.76	57.40g-i	1.98	95.26c-h	2.15	141.87c-h
CML 495 x CML 343	0.06	1.14a	1.90	79.63b-g	1.46	28.82a-c
CL-07304 x CML448	3.05	1127.98j	.	.	1.85	70.60b-e
CL-07305 x CML448	1.04	10.90a-i	2.20	157.43e-j	1.89	77.71b-f
CL-PSTG01 x CL-04343	0.46	2.86a-e	1.78	60.83a-d	1.72	52.00b-d
CL-RCW73 x CL-04343	-0.05	0.90a	2.16	143.25d-h	2.34	218.07d-i
CL-RCW35 x CL-04343	0.28	1.90ab	1.50	31.46a	1.55	35.88a-c
CL-RCW45 x CL-04343	1.80	62.49g-i	2.30	199.71h-j	2.38	240.27d-i
CL-RCW78 x CL-04343	0.42	2.60a-d	1.97	93.93c-h	1.68	48.18b-d
CML404 x CML476	0.36	2.28a-c	1.96	90.61c-h	0.89	7.79a
CML494 x CML495	0.99	9.80a-i	1.86	71.71a-g	2.00	100.67b-g
CML247 x CML254	0.71	5.11a-g	1.98	96.54c-h	2.99	981.75ij
CLQ-RCWQ15 x CML491	1.65	44.33e-i	2.23	169.36f-j	2.42	265.95d-j
CML 254 x CML 343 (Af Resistant Check)		.	2.07	117.17d-h	1.42	26.41a-c
CML 343 x CML 449 (Af Resistant Check)	0.62	4.20a-g	2.10	126.91d-h	1.49	30.77a-c
(CML448xCML449) x CML450	1.82	65.45g-i	2.00	100.81c-h	2.01	102.19b-g
(CML264QxCML150) x CML491	0.66	4.59a-g	2.24	175.35g-j	3.15	1428.24j
CML311 x (AC7643/AC7729/TZSRW)-1-75-#-BBBB-1-2-6-BB	0.49	3.12a-f	2.17	148.25d-i	2.65	442.59f-j
CML311 x [P44c8FS158-3-2-4-1-BB x CML321]F2-38-1-BB	1.34	21.93b-i	2.24	173.66g-j	2.42	260.02d-j
CML311 x 95SLW HG"B"c1-9-1-4-2-1-B	1.12	13.33a-i	1.81	64.00a-e	2.90	796.53h-j
[CML264 x CML311] x CML334	1.98	94.67h-j	2.15	140.80d-h	2.50	319.60e-j
[CML78 x CML373] x TR res EC/MBR IPTT-ECBMo.88-91-2-1-2-(1-6)#b1 16# 16#B1-1-1-B-#-B*6	0.78	6.08a-h	1.55	35.26ab	1.74	54.76b-d

Table 3.6. Continued

Pedigree	Texas Log AF	Texas Antilog AF	Georgia Log AF	Georgia Antilog AF	Mississippi Log AF	Mississippi Antilog AF
[CML78 x CML373] x TR res EC/MBR IPTT-ECBMo.88-91-2-1-2-(1-6)#b1 16#b 16#b-4-1-B-#-B*11	1.96	91.41h-j	1.84	69.09a-f	1.30	19.93ab
CML373 x CML384	2.15	141.68ij	2.75	558.34k	2.40	249.34e-i
CML311 x CML384	0.70	4.99a-g	2.56	360.66i-k	2.90	785.60h-j
CML 491 x CML 150 (Af Susceptible Check)	1.82	66.15g-i	1.97	93.73c-h	2.95	900.74ij
Wilson 1851W	1.69	49.49f-i	.	.	.	.
RX949W	1.62	41.70d-i	.	.	.	.
RX953W	1.52	33.27c-i	.	.	.	.
Triumph 1910W	1.52	32.98c-i	.	.	.	.
P32H39	1.53	33.64c-i	.	.	.	.
Mp313E x Mo18W	.	.	2.20	157.54e-j	1.73	53.95b-d
SC212M x GA209	.	.	2.56	366.44jk	.	.
T173 x Va35	.	.	2.34	219.03h-j	.	.
Z1851W	.	.	2.59	386.63jk	.	.
Z64W	.	.	2.57	375.23jk	.	.
Mp 313E x Tex 6	.	.	.	.	1.82	66.57b-e
Mp 494 x Mp 717	.	.	.	.	1.57	37.49a-c
GA 209 x Mp 339	.	.	.	.	2.53	341.11e-j
GA 209 x SC 212M	.	.	.	.	2.67	468.17g-j
Mean	**1.16**	**28.02**	**2.09**	**156.35**	**2.10**	**262.29**
MSE	**0.31**	**.**	**0.07**	**.**	**0.20**	**.**
LSD	**1.21**	**.**	**0.39**	**.**	**0.76**	**.**
Sig	**0.00**	**.**	**0.00**	**.**	**0.00**	**.**
Min	**0.05**	**0.90**	**1.50**	**31.46**	**0.89**	**7.79**
Max	**3.05**	**141.68**	**2.75**	**558.34**	**3.15**	**1428.24**

Conclusions

Individual inbred lines, yellow hybrids, and white hybrids accumulated significantly less aflatoxin than US checks. These lines have potential for use in areas where aflatoxin is a chronic problem. Further evaluation is needed to determine how well these maize lines and hybrids perform across years. Yield trials are also needed to ensure aflatoxin tolerant hybrids have acceptable grain yield.

The effect of environment and genotype by environment interaction was significant in all three experiments. There are few genotypes included in this study that showed a similar response in each location. The most stable lines tended to be consistently average or poor performing. The most promising lines appear to be adapted to a specific environment, accumulating low levels of aflatoxin in one location and greater amounts in the other locations tested. These lines must prove to be stable across years to be useful. The difference in inoculation method between locations may have contributed to the variation between environments.

CHAPTER IV

GRAIN COMPOSITION

Introduction

Corn is grown primarily as a carbohydrate source and from this basis there are a myriad of potential markets for the grain and its derivatives. While the majority of corn production is used as a feed grain or ethanol feedstock, the milling industry demands a sizable portion of the corn production as well. The majority of corn that is milled is wet-milled, which divides the grain into chemical components. Maize is an important raw material in the starch industry because it is rich in starch and the starch extraction process yields useful byproducts (Haros and Suarez, 1997). The wet-milling industry provides a stable demand for maize grain, enhancing long-term profitability of producers (Zehr et al., 1996). A market is developing for high-starch maize (Eckhoff, 1995; Paulsen et al., 2003).

Maize hybrids with increased starch content are believed to have greater end-use values (Fox et al., 1992). A 1% increase in starch content is worth $0.02 to $0.03 per bushel (Eckhoff, 1995; Eckhoff et al., 1996; Paulsen et al., 2003) based on the difference in the value of starch and the by-product gluten feed. Although the value is difficult to quantify, higher quality maize is usually worth about $0.15 to $0.20 per bushel when increased process efficiency and better capital utilization is added (Eckhoff, 1995). Low starch yields result in high co-product production and when the capacity of any co-product stream is exceeded, the grind rate must be decreased (Eckhoff, 1995). Variation in grind characteristics results in reduced milling efficiency (Zehr et al., 1995).

Grain is fractioned into products representing its major components, starch, protein, oil, and fiber through the wet-milling process (Zehr et al., 1996). Starch is the major product of the wet-milling process in which the maize is steeped in an aqueous sulphurous acid solution to facilitate the separation of the germ, hull, gluten, and starch (Haros and Suarez, 1997). In addition to starch, wet-milling also generates useful by-products including 21% protein gluten feed and 60% protein gluten meal for livestock feed, corn oil, and steep liquor used for pharmaceutical fermentations (Eckhoff, 1997).

Starch recovered by the wet-milling process is converted into many food and industrial products (Zehr et al., 1996). The three largest uses for corn starch include corn sweeteners, ethanol, and starch. Other uses include paper use, cereals, alkaline cooked food products, and adhesives (Orman and Schumann, 1991; Paulsen et al., 1996; Eckhoff, 1997).

High starch yield is dependent on maize variety, environmental conditions, and drying conditions (Paulsen et al., 1996, 2003). The specific maize hybrid is the most important factor for starch yield (Brumm et al., 1991; Zehr and Eckhoff, 1995; Haros and Suraez, 1997; Singh et al., 1998). Nearly half of the variation in starch yield was accounted for among hybrids (Fox et al., 1992). Another study attributed greater than 70% of the variation in starch yield to differences among genotypes. Hybrids with a larger kernel size have shown increased starch yield (Zehr et al., 1995). In addition to genetic effects, environmental conditions including a long growing season, cool evenings, and no early frost aid in good kernel fill and high starch yields (Paulsen et al., 1996).

Analysis of wet-milling properties in grain requires relatively large samples and is time and labor intensive so it has not been used in large scale genetic studies (Zehr et al., 1996). While difficult, corn improvement programs have been successful in modifying grain composition because product fractions from wet-milling are heritable and can be modified through selection (Zehr et al., 1996). More progress could be expected if simpler and faster methods to predict starch yield were developed to aid in selecting cultivars in a breeding program and for identifying maize shipments that have desirable processing characteristics (Wehling et al., 1993).

One potential method is to use near-infrared spectroscopy (NIR) as this method can provide whole kernel analysis of starch, moisture, protein, fiber, and oil percentages in less than a minute (Paulsen et al., 2003). NIR is a widely used analytical method used by the grain industry to determine kernel composition (Wehling et al., 1993). It is a non-destructive sampling procedure so seed analyzed for grain composition remains viable for planting. One significant limitation of NIR is that it measures starch content rather than starch yield (Paulsen et al., 2003).

Starch content measured by NIR was highly correlated to laboratory wet-milling procedures (r=.80) in a study of 200 maize genotypes (Dijkhuizen, 1998). The NIR method is suitable to aid in selection of increased starch content in maize, (Zehr and Eckhoff, 1995; Zehr et al., 1996; Dijkhuizen, 1998; Paulsen, 2003) but may not alone be adequate to predict final relative wet-milling performance among genotypes (Zehr et al., 1996).

An average sample of maize grain would consist of 71% starch, 9% protein, and 4% oil on a dry weight basis (Orman and Schumann, 1991). Commercial hybrid samples vary by 10-18% in starch yield (Eckhoff et al., 1996). Studies have reported different ranges for starch yield:

- 46.5-62.3% (Zehr et al., 1995)
- 47-72% (Echhoff, 1995)
- 47.2-64.5% (Wehling et al., 1993)
- 54-72% (Eckhoff, 1996, 1997)
- 57.6-60.6% (Fox et al., 1992)
- 58-72% (Paulsen et al., 2003)
- 62-70% (Paulsen et al., 2003)
- 66-71% (Paulsen et al., 1996)
- 67.4-75.8% (Orman and Schumann, 1991)

Higher starch yielding genotypes generally have less starch in the fiber, higher protein gluten meal, and cleaner germ when it is wet-milled (Eckhoff, 1997). There is a general tendency for varieties high in starch to be low in protein (Paulsen et al., 1996). The Illinois high protein maize and Illinois low protein maize populations clearly demonstrate this trend. Starch yields ranged from 39.0% in the high protein population to 68.8% in the low protein population (Paulsen et al., 2003).

Given this background, the objective of this experiment is to compare starch content of exotic adapted hybrids, advanced yellow testcrosses, and quality protein maize hybrids to US hybrid maize.

Materials and Methods

Entries for this experiment were selected primarily from the collection of LAMA testcrosses included in the aflatoxin trials reported in chapter II. These LAMA lines are 100% tropical adapted lines originating in Latin America. US hybrids were included as checks representing temperate US maize. Other genotypes including quality protein maize (QPM) lines and advanced yellow maize lines from the Texas A&M University maize breeding program were also included to compare the starch content of different types of maize lines. All hybrids were developed by the Corn Breeding and Genetics program at Texas A&M University, seed was produced in College Station 2004 summer nursery and Weslaco 2005 winter nursery.

All 54 hybrid entries were grown in replicated field trials in College Station in 2006 and in Weslaco in the 2006 winter nursery. The plots were irrigated and managed for maximum yield production. In College Station, the hybrids were open pollinated and three random ears from each plot were harvested by hand. In Weslaco the hybrids were selfed and all selfed ears, 8-10 ears per plot, were harvested. The grain from the Weslaco trial was divided into two sub-samples per plot.

Starch measurements were taken using an Infratec 1226 Grain Analyzer NIR intrument. The NIR reports protein and oil content along with starch content so all three traits were recorded and analyzed. The NIR was programmed to measure two sub-samples of each grain sample from the College Station trial, a larger grain sample was available from the Weslaco trial so three sub-samples were measured. Each grain sample was run through the NIR three times so a mean and standard deviation for each

measurement could be calculated. Statistical analysis was conducted using the SAS proc GLM procedure.

Results and Discussion

In the College Station trial, starch content ranged from a minimum of 69.08 percent to a maximum of 72.48 (Table 4.1). The entries Tx732-B-B-B/LH210 (72.48%), Temp. NSSLate B-103-B-2-B-B-B-B/LH195 (72.36%), and LAMA2002-37-2-B-B-B/LH195 (72.34%) had the highest starch content in the trial while LAMA2002-8-1-B-B-B/LH195 (69.08%), ((Ko326y x Tx806)-6-1-1-1-B-B/CML161)x(Tx802/CML161))-2-B-B-B-B-2/LH195 (69.45%), and Temperate x Tropical High-Oil QPM-B-5-B-1-B-B-B-B-B/LH195 (69.65%) had the lowest starch content. Protein measurements showed a wider range, with a minimum of 7.80% and a maximum of 12.01%. Oil content was the least variable trait with a range of 1.14 percent. There were significant differences between genotypes for oil, protein, and starch content (Table 4.2). Measurement was not significant indicating that the NIR method is precise for measuring starch, oil, and protein content in whole kernel corn samples.

Table 4.1. Mean oil, protein, and starch content of entries in the College Station hybrid trial in 2006.

Pedigree	Material	oil (%)	protein (%)	starch (%)
LAMA2002-10-1-B-B-B/LH195	Exotic[a]	3.76	11.00	70.80
LAMA2002-10-2-B-B-B/LH195	Exotic	3.89	10.65	70.71
LAMA2002-12-1-B-B-B/LH195	Exotic	3.77	10.66	70.55
LAMA2002-1-2-B/LH195	Exotic	3.78	8.55	71.93
LAMA2002-13-B2-B-B-B/LH195	Exotic	4.45	10.09	69.90
LAMA2002-14-B-B/LH195	Exotic	4.29	10.31	69.96
LAMA2002-1-5-B-B-B/LH195	Exotic	3.81	9.59	71.56
LAMA2002-20-2-B-B-B/LH195	Exotic	4.21	10.63	70.15
LAMA2002-20-4-B/LH195	Exotic	3.10	10.11	70.49
LAMA2002-20-4-B/LH210	Exotic	3.76	8.78	71.77
LAMA2002-2-1-B/LH210	Exotic	4.24	8.77	71.19
LAMA2002-22-1-B-B-B/LH195	Exotic	3.88	9.87	70.63
LAMA2002-22-3-B/LH195	Exotic	3.48	12.01	69.96
LAMA2002-22-3-B/LH210	Exotic	3.68	10.74	70.47
LAMA2002-22-3-B-B2-B/LH195	Exotic	3.57	11.09	70.69
LAMA2002-2-3-B/LH210	Exotic	3.98	9.35	71.31
LAMA2002-25-4-B-B-B/LH195	Exotic	3.62	9.94	71.14
LAMA2002-25-5-B/LH195	Exotic	4.17	9.38	70.19
LAMA2002-2-6-B/LH195	Exotic	3.77	9.81	71.04
LAMA2002-2-6-B/LH210	Exotic	3.75	9.55	71.41
LAMA2002-27-1-B-B-B/LH195	Exotic	3.98	9.93	70.93
LAMA2002-35-2-B-B/LH195	Exotic	3.87	10.05	71.02
LAMA2002-37-2-B-B-B/LH195	Exotic	3.43	9.18	72.33

Table 4.1. Continued

Pedigree	Material	oil (%)	protein (%)	starch (%)
LAMA2002-44-B-B-B-B/LH195	Exotic	3.92	9.71	70.97
LAMA2002-46-3-B-B-B/LH195	Exotic	4.03	11.34	70.00
LAMA2002-46-6-B/LH195	Exotic	4.29	9.41	70.60
LAMA2002-46-6-B/LH210	Exotic	4.05	8.90	71.36
LAMA2002-53-1-B/LH195	Exotic	4.46	9.07	70.67
LAMA2002-53-5-B/LH195	Exotic	3.86	9.62	71.07
LAMA2002-53-5-B/LH195	Exotic	3.63	9.83	71.22
LAMA2002-53-5-B/LH210	Exotic	4.44	8.64	70.91
LAMA2002-58-3-B-B-B/LH195	Exotic	4.05	10.46	70.40
LAMA2002-58-7-B/LH195	Exotic	3.97	9.90	70.86
LAMA2002-61-2-B/LH195	Exotic	3.83	10.32	70.46
LAMA2002-61-6-B-B-B/LH195	Exotic	3.87	10.23	70.91
LAMA2002-8-1-B-B-B/LH195	Exotic	4.08	11.97	69.07
LAMA2002-9-3-B/LH195	Exotic	3.95	9.66	70.91
LAMA2002-9-3-B/LH210	Exotic	3.98	9.17	70.95
(B104-1 x Tx714-B/B110 x FR2128-B)-12-4-B-B-B-B/LH210	Yellow[b]	3.86	8.27	71.50
(B102-1 x NC300/B100 x FR2128)-3-1-B1-B-B-B/LH210	Yellow	3.79	9.46	71.55
(B104-1 x Tx714-B-B)-9-1-B-B-B-B/LH210	Yellow	3.83	7.92	72.04
(B104-1 x Tx714-B-B)-17-2-B-B-B-B/LH210	Yellow	3.88	9.42	71.56
Tx732-B-B-B/LH210	Yellow	3.84	7.80	72.48
FRB73-B-B/LH210	Yellow	3.97	9.10	71.17
Tx759 (Tx6252/Va35)-1-1-2-2-3-6-1-B-B-B-B-B/LH210	Yellow	4.18	9.36	70.74
((B104/NC300)x(CML 415/B104))-4-2-B-B-B/LH287RR	Yellow	3.74	8.93	71.43

Table 4.1. Continued

Pedigree	Material	oil (%)	protein (%)	starch (%)
Temperate x Tropical High-Oil QPM-B-5-B-1-B-B-B-B-B/LH195	QPM[c]	4.53	10.57	69.64
Temp. NSSLate B-103-B-2-B-B-B-B/LH195	QPM	3.85	7.83	72.39
DKC66-80	US[d]	3.86	8.26	72.15
DKC66-80	US	3.76	8.68	72.08
DKC69-70	US	3.45	9.07	71.61
DKC69-71	US	3.69	9.41	70.95
DKC69-71	US	3.51	9.87	71.40
DKC69-71	US	3.70	10.19	70.58
P31B13	US	3.74	8.97	71.62
P31B13	US	4.06	8.83	70.81
P31B13	US	3.69	8.60	71.89
P31G66	US	3.64	7.89	72.59
W4700	US	4.52	9.25	71.77
Mean		**3.95**	**9.75**	**70.88**
MSE		**0.16**	**0.30**	**0.59**
LSD		**0.46**	**0.62**	**0.87**
Sig		**0.00**	**0.00**	**0.00**
Min		**3.43**	**7.80**	**69.07**
Max		**4.55**	**12.01**	**72.48**

[a] 100% exotic adapted lines testcrossed to LH195 or LH210
[b] Texas A&M University corn breeding program advanced yellow hybrid
[c] Texas A&M University corn breeding program advanced QPM testcross with LH195
[d] US hybrid

Table 4.2. ANOVA for oil, protein, and starch contents for College Station trial in 2006.

		oil		protein		starch	
Source	df	mean square	Pr>F	mean square	Pr>F	mean square	Pr>F
Entry	63	0.522461	<.0001	5.947490	<.0001	3.743188	<.0001
Rep	2	0.475007	0.0541	3.019654	0.0008	0.089784	0.8793
Measurement	2	0.054324	0.7141	0.017465	0.9582	0.064398	0.9119
Entry*Measurement	124	0.163954	0.4470	0.057401	1.0000	0.362976	1.0000
Error	270	0.161105		0.408908		0.697941	
R-square		0.557569		0.777886		0.599979	

The Weslaco hybrid trial (Table 4.3) has higher mean oil, protein, and starch content than the College Station trial. This difference may be due to differences in grain moisture content rather than real differences in grain composition, but moisture content was not measured. The oil content in the Weslaco trial ranged from 3.43 to 4.88 percent. Protein content ranged from 8.72 to 13.01 percent, and starch content ranged from 69.93 to 73.90 percent. Advanced yellow testcross (B102-1 x MC300/B100 x FR2128)-3-1-B1-B-B-B/LH210 and exotic adapted testcrosses LAMA2002-27-1-B-B-B/LH195 and LAMA2002-22-3-B-B2-B/LH195 were the trial entries with the highest starch content. Exotic adapted testcrosses LAMA2002-8-1-B-B-B/LH195, LAMA2002-37-2-B-B-B/LH195, and LAMA2002-12-1-B-B-B/LH195 had the lowest starch content.

Table 4.3. Mean oil, protein, and starch content of entries in the Weslaco hybrid trial in 2006.

Pedigree	Material	Oil (%)	Protein (%)	Starch (%)
LAMA2002-1-5-B-B-B/LH195	Exotic[a]	4.32	10.48	71.89
LAMA2002-2-3-B/LH210	exotic	4.16	9.68	72.15
LAMA2002-2-6-B/LH195	exotic	4.29	9.82	72.39
LAMA2002-8-1-B-B-B/LH195	exotic	4.50	13.01	69.93
LAMA2002-9-2-B/LH210	exotic	4.45	9.12	72.15
LAMA2002-9-3-B/LH195	exotic	3.94	12.81	70.34
LAMA2002-9-3-B/LH210	exotic	4.27	9.93	72.11
LAMA2002-10-1-B-B-B/LH195	exotic	4.59	11.53	70.66
LAMA2002-10-2-B-B-B/LH195	exotic	4.37	11.46	70.89
LAMA2002-12-1-B-B-B/LH195	exotic	4.86	11.59	70.18
LAMA2002-13-B2-B-B-B/LH195	exotic	4.48	11.46	70.70
LAMA2002-20-2-B-B-B/LH195	exotic	4.88	11.56	70.35
LAMA2002-20-4-B/LH210	exotic	4.17	10.33	71.80
LAMA2002-22-1-B-B-B/LH195	exotic	4.15	10.98	71.41
LAMA2002-22-3-B/LH195	exotic	3.84	12.32	70.76
LAMA2002-22-3-B-B2-B/LH195	exotic	3.77	8.89	73.90
LAMA2002-23-3-B/LH195	exotic	4.04	11.72	70.88
LAMA2002-25-4-B-B-B/LH195	exotic	4.34	11.34	70.97
LAMA2002-25-5-B/LH195	exotic	4.21	11.61	70.36
LAMA2002-27-1-B-B-B/LH195	exotic	3.87	9.16	73.82
LAMA2002-34-7-B/LH195	exotic	4.64	10.70	71.19
LAMA2002-35-2-B-B/LH195	exotic	3.72	11.96	71.24
LAMA2002-37-2-B-B-B/LH195	exotic	4.71	11.81	70.14
LAMA2002-42-B-B-B-B/LH195	exotic	4.53	11.46	70.48
LAMA2002-46-3-B-B-B/LH195	exotic	3.91	11.72	71.22
LAMA2002-46-6-B/LH195	exotic	4.44	11.81	70.34
LAMA2002-46-6-B/LH210	exotic	4.21	8.81	73.12
LAMA2002-58-3-B-B-B/LH195	exotic	4.36	11.87	70.56
LAMA2002-61-2-B/LH195	exotic	3.92	9.40	72.69

Table 4.3. Continued

Pedigree	Material	Oil (%)	Protein (%)	Starch (%)
LAMA2002-61-6-B-B-B/LH195	exotic	3.99	12.01	70.76
((B104/NC300)x(CML 415/B104))-4-2-B-B-B/LH287RR	Yellow[b]	3.90	9.31	73.16
((Ko326y x Tx806)-6-1-1-1-B-B/CML161)x(Tx802/CML161))-2-B-B-B-B-1/LH195	QPM[c]	4.76	11.23	70.44
((Ko326y x Tx806)-6-1-1-1-B-B/CML161)x(Tx802/CML161))-2-B-B-B-B-2/LH195	QPM	4.39	11.70	70.41
Pop. 69 Templado Amarillo QPM-B-B-B6-8-B-B-B-B-B/LH195	QPM	4.50	11.08	70.53
Temperate x Tropical High-Oil QPM-B-5-B-1-B-B-B-B-B/LH195	QPM	4.21	12.45	71.04
Temp. NSSLate B-103-B-2-B-B-B-B/LH195	QPM	4.03	10.16	71.29
(B104-1 x Tx714-B/B110 x FR2128-B)-12-4-B-B-B-B/LH210	Yellow	3.91	10.07	71.94
(B102-1 x NC300/B100 x FR2128)-3-1-B1-B-B-B/LH210	Yellow	3.87	8.72	73.56
(B104-1 x Tx714-B-B)-9-1-B-B-B-B/LH210	Yellow	3.73	9.95	72.50
(B104-1 x Tx714-B-B)-17-2-B-B-B-B/LH210	Yellow	3.48	9.47	73.06
Tx732-B-B-B/LH210	Yellow	3.62	10.09	72.69
FRB73-B-B/LH210	Yellow	3.44	9.72	72.90
Tx759 (Tx6252/Va35)-1-1-2-2-3-6-1-B-B-B-B-B/LH210	Yellow	4.37	9.23	72.30
DKC69-70	US[d]	3.95	11.70	70.44
DKC69-71	US	4.43	10.91	70.88
DKC66-80	US	4.49	10.76	71.06
P31B13	US	4.01	10.24	72.23
W4700	US	3.43	10.66	71.82
Mean		**4.18**	**10.73**	**71.54**
MSE		**0.05**	**0.09**	**0.25**
LSD		**0.30**	**0.39**	**0.65**
Sig		**0.00**	**0.00**	**0.00**
Min		**3.43**	**8.72**	**69.93**
Max		**4.88**	**13.01**	**73.90**

[a] 100% exotic adapted lines testcrossed to LH195 or LH210
[b] Texas A&M University corn breeding program advanced yellow hybrid
[c] Texas A&M University corn breeding program advanced QPM testcross with LH195
[d] US hybrid

Significant differences were observed among genotypes for oil, protein, and starch content in the Weslaco trial (Table 4.4). Measurement was not significant for starch or oil, but was significant for protein content. That significant difference for protein content measurement indicates a limitation of the NIR method for measuring protein content due to the lack of precise measurements although it was able to measure significant differences between genotypes. This method can rank genotypes based on protein content but, due to measurement error, cannot give a precise value of protein content for each genotype.

Table 4.4. ANOVA for oil, protein, and starch contents in the Weslaco hybrid trial in 2006.

		Oil		Protein		Starch	
Source	df	mean square	Pr>F	mean square	Pr>F	mean square	Pr>F
Entry	49	31.63836629	<.0001	6.7338194	<.0001	6.4208243	<.0001
Measurement	2	0.08777796	0.409	0.1793406	0.0211	0.1292613	0.5359
Entry*Measurement	98	6.0594553	0.1094	0.139338	<.0001	0.3164719	0.0137
Error	114	0.04870088		0.044939		0.2061022	
R-square		0.871656		0.985315		0.936349	

In the combined analysis, significant differences between genotypes were found for protein and starch content, but not for oil content (Table 4.5). The effect of environment and the genotype by environment interaction was significant for all three traits. As observed in individual environments, measurement was not significant for all three traits. This indicates the method had adequate precision for measuring oil, protein, and starch content.

Table 4.5. ANOVA for oil, protein, and starch content in the hybrid trial across locations in 2006.

		Oil		Protein		Starch	
Source	df	mean square	Pr>F	mean square	Pr>F	mean square	Pr>F
Genotype	61	0.65495661	0.1575	8.4672024	0.0012	6.3794157	0.0071
Environment	1	5.23245209	<.0001	131.8783603	<.0001	44.2460765	<.0001
Measurement	2	0.02596375	0.8113	0.0139849	0.9411	0.0430081	0.9144
Rep	2	0.50287957	0.0179	2.0879064	0.0001	0.2593504	0.5832
GEI	46	0.49266257	<.0001	3.5339064	<.0001	3.1647292	<.0001
Error	613	0.1241206		0.23017		0.4805984	
R-square		0.489664		0.863246		0.676232	

Conclusions

The NIR method effectively identified differences between genotypes for starch, protein, and oil content. Some of the exotic adapted LAMA lines have significantly higher starch content than the US hybrid checks. However, the LAMA hybrids have a hard, flint type kernel that is undesirable for wet milling because flint-types would require longer steep times which reduce milling efficiency. Although high starch content was observed in exotic adapted LAMA testcrosses, they have little breeding value due to their unsuitability for wet milling. Advanced yellow hybrid Tx732-B-B-B/LH210 had the highest starch content across locations and is more suitable for wet-milling. The grain composition of more lines from the breeding program could be evaluated to identify additional high starch lines. This test did illustrate that the NIR method is effective and precise enough to measure differences between genotypes for starch content. Grain moisture should also be measured in future studies to eliminate error due to differences in grain moisture content.

CHAPTER V

SUMMARY

Exotic maize germplasm is often cited as a source of genes for specific traits. Exotic maize with reduced susceptibility to aflatoxin accumulation and exotic hybrids with high starch content were identified. The strategy of developing exotic adapted lines was effective for the traits of interest, but the resulting lines are still of limited usefulness due to their low grain yield potential.

LAMA Aflatoxin Trials

The LAMA testcrosses consistently accumulated less aflatoxin than US hybrids across years and environments. This collection of exotic adapted testcrosses may contain aflatoxin resistance factors not currently available in US germplasm. The mean grain yield for the LAMA testcrosses was lower than the US hybrids in all environments and locations. In each environment, there were individual testcrosses that yielded competitively with US hybrids. In Bardwell, Weslaco, and Corpus Christi five to ten testcrosses were statistically similar in yield with the highest yielding US hybrid in the 2005 trial. In College Station and Dalhart, only one and two testcrosses, respectively did not yield significantly less than the highest yielding US hybrid while all LAMA testcrosses yielded less than the best yielding US hybrid in Castroville and Wharton. In 2006, 12 testcrosses in Bardwell and 14 LAMA testcrosses in College Station had yield that were statistically similar to the highest yielding US hybrid while all testcrosses were significantly lower yielding in the Weslaco and Wharton trials.

The LAMA testcrosses have higher mean grain moisture and test weight than the US hybrids. To use this germplasm directly in US breeding programs, grain moisture content will need to be reduced.

CIMMYT Aflatoxin Trials

Inbred lines, yellow hybrids, and white hybrids were identified that accumulate less aflatoxin than current US hybrids. Genotype by environment interaction was significant in all three trials indicating that these lines may be better adapted to a narrow environment than the wider range of environments in which they were tested. Additional trials are required to determine the stability of these lines across years in the same environment. Yield trials are also needed to compare the grain production of these yellow and white hybrids to current US hybrids.

Grain Starch Content

LAMA testcross hybrids and advanced yellow testcrosses were identified with higher starch content than US hybrids. The NIR method effectively measured differences between genotypes for starch, oil, and protein content of the grain, confirming the method's suitability for use in early generation analysis in a maize breeding program.

REFERENCES

Albrecht, Bruno, and .J.W. Dudley. 1987. Evaluation of Four Maize Populations Containing Different Proportions of Exotic Germplasm Crop Science 27:480-486.

Betran, F.J., T. Isakeit, and G. Odvody. 2002. Aflatoxin Accumulation of White and Yellow Maize Inbreds in Diallel Crosses. Crop Science 42:1894-1901.

Betran, F.J.and T.Isakeit. 2004. Aflatoxin Accumulation in Maize Hybrids of Different Maturities. Agronomy Journal 96:565-570.

Betran, F.J., K. Mayfield, T. Isakeit, and M. Menz. 2005. Breeding Exotic Maize Germplasm. pp. 353-377. *In* Proceedings of the Arnel R. Hallauer International Plant Breeding Symposium. Mexico D.F., Mexico 2003. Iowa State Press.

Brown, R.L., Z.Y. Chen, T.E. Cleveland, J.S. Russin. 1999. Advances in the Development of Host Resistance in Corn to Aflatoxin Contamination by *Aspergillus flavus*. Phytopathology 89: 113-117.

Brown, W.L. 1975. A Broader Germplasm Base in Corn and Sorghum. Proc. Annual Corn Sorghum Res. Conf. 30: 81-89.

Brumm, T.J., L.A. Johnson, C.R. Hurburgh. 1991. The Potential of Near-infrared Spectroscopy for Determining Maize Wet Milling Characteristics. Cereal Foods World 36:706.

Campbell, K.W.and D.G.White. 1995a. Evaluation of Corn Genotypes for Resistance to Aspergillus Ear Rot, Kernel Infection, and Aflatoxin Production. Plant Disease 79:1039-1045.

Campbell, K.W.and D.G.White. 1995b. Inheritance of Resistance to Aspergillus Ear Rot and Aflatoxin in Corn Genotypes. Phytopathology 85:886-896.

Cleveland, T.E., P.F. Dowd, A.E. Desjardins, D. Bhatnagar, P.J. Cotty. 2003. United States Department of Agriculture - Agriculture Research Service Research on Pre-harvest Prevention of Mycotoxins and Mycotoxigenic Fungi in U.S. crops. Pest Management Science 59:629-642.

Darrah, L.L., E.B. Lillehoj, M.S. Zuber, G.E. Scott, D. Thompson, D.R. West, N.W. Widstrom, and B.A. Fortnum. 1987. Inheritance of Aflatoxin B1 Levels in Maize Kernels under Modified Natural Inoculation with *Aspergillus flavus*. Crop Science 27:869-872.

Dijkuizen, A.J.W.D., T.R. Rocheford, A. E. Haken, S.R. Eckhoff. 1998. Near-Infrared Reflectance Correlated to 100-g Wet-Milling Analysis in Maize. Cereal Chemistry 75:266-270.

Duvick, D.N. 1975. Using Host Resistance to Manage Pathogen Populations Iowa State Journal of Research 49:505-512.

Duvick, D.N. 1981. Genetic Diversity in Corn Improvement. Annual Corn Sorghum Research Conference Proc. 36: 48-60.

Echandi, C.R.and A.R.Hallauer. 1996. Evaluation of U.S. Corn Belt and Adapted Tropical Maize Cultivars and Their Diallel Crosses. Maydica 41:317-324.

Eckhoff, S.R. 1995. The Future of Commodity Maize. Wet Milling Notes. No. 11, Univ. of Illinois, Agricultural Engineering Dept., Urbana-Champaign, IL.

Eckhoff, S.R., K. Singh, B.E. Zehr, K.D. Rausch, E.J. Fox, A.K. Mistry, A.E. Haken, Y.X. Niu, S.H. Zou, P. Buriak, M.E. Tumbleson, P.L. Keeling. 1996. A 100-g Laboratory Maize Wet Milling Procedure. Cereal Chemistry 73:54-57.

Eckhoff, S.R. 1997. Grain Quality Issues Related to Corn Wet Milling. Agricultural Engineering Department, University of Illinois, Urbana-Champaign, IL.

Fox, S.R., L.A. Johnson, C.R. Hurburgh Jr., C. Dorsey-Redding, T.B. Bailey. 1992. Relations of Grain Proximate Composition and Physical Properties to Wet-Milling Characteristics of Maize. Cereal Chemistry 69:191-197.

Gardner, C.A.C., L.L. Darrah, M.S. Zuber, and J.R. Wallin. 1987. Genetic Control of Aflatoxin Production in Maize. Plant Disease 71:426-429.

Geadelmann, J. 1984. Using Exotic Germplasm to Improve Northern Corn. pp. 98-110. *In* 39th Annual Corn and Sorghum Research Conference. ASTA, Washington, D.C.

Goodman, M.M. 1985. Exotic Maize Germplasm: Status, Prospects, and Remedies. Iowa State Journal of Research:497-527.

Goodman, M.M., F. Castillo, and R.N. Holley. 1988. US Maize Germplasm: Origin, Limitations, and Alternatives. Pp 130-148. *In*: Russell N. and G.M. Listman (eds.) Recent Advances in the Conservation and Utilization of Genetic Resources: Proc. Global Maize Germplasm Workshop. CIMMYT, Mexico, DF.

Goodman, M.M., F. Castillo, and J. Moreno. 1990. Choosing and Using Exotic Maize Germplasm. Annual Illinois Corn Breeders School Proc. pp. 148-171.

Goodman, M.M. 1992. Choosing and Using Tropical Corn Germplasm. Ann. Corn Sorghum Ind. Res. Conf. Proc. 47: 47-64.

Gorman, D.P., M.S. Kang, T. Cleveland, and R.L. Hutchinson. 1992. Combining Ability for Resistance to Field Aflatoxin Accumulation in Maize Grain. Plant Breeding 109:292-303.

Guo, B.Z., J.S. Russin, T.E. Cleveland, R.L. Brown, N.W. Widstrom. 1995. Wax and Cutin Layers in Maize Kernels Associated with Resistance to Aflatoxin Production by *Aspergillus flavus* Journal of Food Protection 58:296-300

Haros, M. and C. Suraez. 1997. Effect of Drying, Initial Moisture, and Variety in Maize Wet Milling. Journal of Food Engineering 34:473-481.

Jones, R.K. 1987. The Influence of Cultural Practices on Minimizing the Development of Aflatoxin in Field Maize. pp 136-144 *In*: Aflatoxin in maize: Proc. Workshop. Zuber, M.S., E.B. Lillehoj, and B.L. Renfro (eds.) International Maize and Wheat Improvement Center, Mexico, DF.

Lewis, R.S., M.M. Goodman. 2003. Incorporation of Tropical Maize Germplasm into Inbred Lines Derived from Temperate x Temperate-adapted Tropical Line Crosses: Agronomic and Molecular Assessment. Theoretical and Applied Genetics 107:798-805.

McMillian, W.W., D.M. Wilson, N.W. Widstrom. 1985. Aflatoxin Contamination of Preharvest Corn in Georgia: A Six-Year Study of Insect Damage and Visible *Aspergillus flavus*. Journal of Environmental Quality 14:200-202.

Michelini, L.A. and A.R.Hallauer. 1993. Evaluation of Exotic and Adapted Maize (*Zea mays* L.) Germplasm Crosses. Maydica 38:275-282.

Moreno, O.J. and M.S.Kang. 1999. Aflatoxins in Maize: The Problem and Genetic Solutions. Plant Breeding 118:1-16.

Munkvold, G.P. 2003. Cultural and Genetic Approaches to Managing Mycotoxins in Maize. Annual Reviews of Phytopathology 41:99-116.

NASS. 2005. Crop Production: 2004 Summary. Crop Production Summary. United States Department of Agriculture – National Agricultural Statistics Service. Washington, D.C.

Ochs, B.A. 2005. Evaluation of Argentine Maize Hybrids and Exotic x Temperate Testcrosses Across Environments. M.S. thesis. Texas A&M University. College Station, TX, USA.

Odvody, G.N., C.F. Chilcutt, R.D. Parker, and J.H. Benedict. 2000. Aflatoxin and Insect Response of Near-isogenic Bt and Non-Bt Commercial Corn Hybrids in South Texas. p. 121-122. *In* Proc. Of the Aflatoxin/Fumonisin Workshop 2000, Yosemite, CA. 25-27 Oct 2000.

Orman, B.A., R.A. Schumann. 1991. Comparison of Near-infrared Spectroscopy Calibration Methods for the Predicting of Protein, Oil, and Starch in Maize Grain. Journal of Agricultural Food Chemistry 39:883-886.

Park, D.L. and B.Liang. 1993. Perspectives on Aflatoxin Control for Human Food and Animal Feed. Trends in Food Science and Technology 4:334-342.

Patterson, H.D. and E.R. Williams. 1976. A New Class of Resolvable Incomplete Block Designs. Biometrica 63: 83-89.

Paulsen, M.R., S.L. Hofing, L.D. Hill, S.R. Eckhoff. 1996. Corn Quality Characteristics for Japan Markets. Applied Engineering in Agriculture 12:731-738.

Paulsen, M.R., L.O. Pordesimo, M.Singh, S.W. Mbuvi, B. Ye. 2003. Maize Starch Yield Calibrations with Near Infrared Reflectance Biosystems Engineering 85:455-460.

Payne, G.A. 1992. Aflatoxin in Maize. Critical Reviews in Plant Sciences 10(5):423-440.

Scott, G.E., N. Zummo. 1988. Sources of Resistance in Maize to Kernel Infection by *Aspergillus flavus* in the Field. Crop Science 28:504-507.

Simmonds, N.W. 1993. Introgression and Incorporation. Strategies for the Use of Crop Genetic Resources. Biology Reviews 68:539-562.

Singh, V., A. Haken, M.R. Paulsen, S.R. Eckhoff. 1998. Starch Yield Sensitivity of Maize Hybrids to Drying Temperature and Harvest Moisture Content. Starch/Starke 50:181-183.

Smith, J.S.C. 1988. Diversity of United States Hybrid Maize Germplasm; Isozyme and Chromatographic Evidence. Crop Science 28:63-69.

Smith, J.S.C., D.N. Duvick, O.S. Smith, A. Grunst, and S.J. Wall. 1999. Effect of Hybrid Breeding on Genetic Diversity in Maize. pp. 119-126. *In*: J.G. Coors and S.

Pandey (eds.) The Genetics and Exploitation of Heterosis in Crops. ASA, Madison, WI.

Stuber, C.W. 1978. Exotic Sources for Broadening Genetic Diversity in Corn Breeding Programs. Ann. Corn Sorghum Ind. Res. Conf. Proc. 33: 34-47.

Tallury, S.P. and M.M.Goodman. 1999. Experimental Evaluation of the Potential of Tropical Germplasm for Temperate Maize Improvement. Theoretical and Applied Genetics 98:54-61.

Uhr D.V. and M.M. Goodman. 1995. Temperate Maize Inbreds from Tropical Germplasm. I. Testcross Yield Trials. Crop Science 35:779-784.

United States Department of Agriculture. 1994. Action Levels for Aflatoxins in Animal Feeds. FDA Compliance Policy Guide. Sec 683.100: 384-385.

Wehling, R.L., D.S. Jackson, D.G. Hooper, A.R. Ghaedian. 1993. Prediction of Wet-milling Starch Yield from Corn by Near-infrared Spectroscopy. Cereal Chemistry 70:720-723.

Widstrom, N.W., D.M. Wilson, and W.W. McMillian. 1981. Aflatoxin Contamination of Preharvest Corn as Influenced by Timing and Method of Inoculation. Applied Environ. Microbiol. 42: 249-251.

Windham, G.L., W.P. Williams, and F.M. Davis. 1999. Effects of the Southwestern Corn Borer on *Aspergillus flavus* Kernel Infection and Aflatoxin Accumulation in Maize Hybrids. Plant Disease 83:535-540.

Windham, G.L., W.P. Williams. 2002. Evaluation of Corn Inbreds and Advanced Breeding Lines for Resistance to Aflatoxin Contamination in the Field. Plant Disease 86:232-234.

Zehr, B.E., S.R. Eckhoff, S.K. Singh, P.L. Keeling. 1995. Comparison of Wet-milling Properties among Maize Inbred Lines and Their Hybrids. Cereal Chemistry 72:491-497.

Zehr, B.E. and S.R. Eckhoff. 1995. Genetic Factors Influencing Wet-milling Quality of Corn. Corn and Sorghum Ind. Res. Conf. Proc. 50: 98-112.

Zehr, B.E., S.R. Eckhoff, W.E. Nyqyist, P.L. Keeling. 1996. Heritability of Product Yields from Wet-milling Maize Grain. Crop Science 36:1159-1165.

APPENDIX

Table A.1. Description of US inbred and hybrid checks included in the CIMMYT aflatoxin trials.

US Check		Description
Mp313E	inbred	Direct self from Tuxpan selected in Mississippi. Long, tight husks, tall plants, high ear placement, late maturity good general combining ability for yield. Resistance to kernel infection by *A. flavus*.
T173	inbred	Selected in Tennessee. White cob and kernels, good to excellent combining ability with BSSS.
Mp717	inbred	Selected in Mississippi. Source of resistance to *A. flavus*. Yellow kernels on a white cob.
Mp 04:96	inbred	Selection in Mississippi from a group of S4's in Antigua Gpo. 2 population. Orange kernels.
Mp715	inbred	Selected for reduced aflatoxin accumulation in Mississippi, developed from Tuxpan. Dark yellow kernels on a white cob.
GA209	inbred	Selection in Georgia from T61 x NC37. White kernels and cob, dent type kernels, good general combining ability.
Mo19W	inbred	Developed in Missouri from WF9/Mo22. White endosperm and cob.
Mp339	inbred	Developed in Mississippi from T61 x Hill Yellow Dent. White kernels and cob.
DKC 69-71	hybrid	Very good disease package for Southern corn producers.
DKC 69-70	hybrid	Good standability, test weight, and quality.
Wilson 1851W	hybrid	White kernels, 116 days to maturity.
Triumph 1910W	hybrid	White kernels

Table A.2. Description of CML lines included in the CIMMYT inbred and hybrid Aflatoxin trials.

CML No.	Adaptation / Program	Maturity	Grain Color	Grain Texture	QPM	Stress tolerance and resistance
78	Subtropical	Interm	W	SD	No	Lodging
144	Lowland	Late	W	F	Yes	
150	Lowland	Late	W	D	Yes	
159	Lowland	Late	W	D	Yes	
161	Lowland	Late	Y	F	Yes	
163	Lowland	Late	Y	D	Yes	
172	Lowland	Late	Y	F	Yes	
176	Subtropical		W	F	Yes	
247	Lowland	Late	W	SD	No	
254	Lowland		W	SD	No	
264	Lowland		W	F	No	
269	Lowland		W	F	No	
285	Lowland		Y	D	No	
287	Lowland		Y	F	No	
311	Subtropical	Late	W	SF	No	
334	Subtropical	Early	W	SD	No	
341	Lowland		W	SD	No	Drought, Low N
342	Lowland		W	SD	No	Drought, Low N
343	Lowland		W	SF	No	Drought
344	Lowland		W	SF	No	Drought, Low N
373	Subtropical	Interm	W	D	No	
384	Subtropical	Late	W	F	No	
404	Lowland	Late	W	F	No	
448	Lowland	Late	W	D	No	
449	Lowland	Interm	W	F	No	
450	Lowland	Late	W	SF	No	
451	Lowland	Late	Y	SF	No	
452	Lowland	Interm	Y	SD	No	
453	Lowland	Late	Y	SD	No	
454	Lowland	Late	Y	F	No	
476	Lowland	Late	W	D	No	
479	Lowland	Interm	Y	F	No	
480	Lowland	Interm	Y	F	No	
481	Lowland	Interm	Y	F	No	
491	Lowland	Late	W	F	Yes	
492	Lowland	Interm	W	F	Yes	
493	Lowland	Interm	Y	SD	Yes	
494	Lowland	Interm	W	D	No	
495	Lowland	Late	W	F	No	

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