INSECT COLONY, PLANTING DATE, AND PLANT GROWTH STAGE EFFECTS ON SCREENING MAIZE FOR LEAF-FEEDING RESISTANCE TO FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE)

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ABSTRACT

Field experiments were conducted at Mississippi State, MS and Tifton, GA to determine effects of laboratory insect colony, planting date, and plant growth stage on screening maize, Zea mays L., for leaf-feeding resistance to the fall armyworm (FAW), Spodoptera frugiperda (J. E. Smith). The experiments were conducted using a randomized complete block design with treatments in a factorial arrangement with 6 replications. Treatments consisted of 2 insect colonies, an early and a late planting period, 2 plant growth stages, and 4 single cross maize hybrids (2 susceptible and 2 resistant to leaf-feeding by FAW) at each location. Each plant in an experiment was infested with 30 neonate FAW larvae when the plants of the second planting within each planting period reached the $\rm V_4$ (Tifton) or $\rm V_8$ (Mississippi State) stage. Each plant was visually scored for leaf damage 7 and 14 days after infestation. Statistical analyses revealed interactions among factors resulting in inferences having to be made using nonmarginal means. Significant differences in rating scores within each factor (insect colony, planting date, and plant growth stage) were found for some comparisons. However, none of these factors appreciably altered our ability to distinguish between resistant and susceptible genotypes which is the objective of screening.

 $\label{eq:continuous} \textbf{Key Words: } Spodoptera\ frugiperda, \ plant\ resistance, \ maize\ hybrids, \ screening$

RESUMEN

Se llevaron a cabo experimentos de campo en el estado de Mississippi y en Tifton, Georgia, para determinar los efectos de una colonia de insectos de laboratorio, fechas de simbra y estado de crecimiento de las plantas en el tamizaje de plantas de maíz, Zea mays L., resistentes al daño causado por Spodoptera frugiperda (J. E. Smith). Los experimentos fueron conducidos usando un diseño de bloques completamente aleatorizados con los tratamientos ordenados en un diseño factorial con 6 réplicas. Los tratamientos consistieron en dos colonias de insectos, un periodo de siembra temprano y otro tardío, dos estados de crecimiento, y cuatro cruces sencillos de maíz híbrido (dos susceptibles y dos resistentes al gusano) en cada localidad. Cada planta en cada experimento fue infestada con 30 larvas neonatas de S. frugiperda cuando las plantas de la segunda siembra dentro de cada período de siembra alcanzaron los estados V. (Tifton) or V_o (Mississippi). Cada planta fue visualmente evaluada en cuanto a daño a las hojas a los 7 y 14 días de la infestación. Los análisis estadísticos revelaron interacciones entre los factores, lo que dió como resutado el tener que hacer inferencias mediante el uso de medias no marginales. En algunas comparaciones fueron encontradas diferencias significativas en el valor de cada factor (colonia de insectos, fecha de siembra, y estado de crecimiento de las plantas). Sin embargo, ninguno de esos factores permitió distinguir entre los genotipos resistentes y susceptibles, lo cual fue el objetivo del tamizaje.

The fall armyworm (FAW), Spodoptera frugiperda (J. E. Smith), is a polyphagous insect that attacks many crops in the Americas (Ashley et al. 1989). In the southeastern United States, the FAW possess a serious threat to maize, Zea mays L., production, especially to late-season plantings (Scott et al. 1977). FAW often attack late-season plantings of maize from seedling to mature stages of growth. Protection of these plantings by multiple applications of insecticides is not practical. Therefore, more economical management tactics must be pursued. The best tactic would be to grow maize that naturally resists the feeding of the pest.

Entomologists and plant geneticists (USDA-ARS) located at Mississippi State, MS and Tifton, GA have screened maize for leaf-feeding resistance to the FAW for many years. Field screening techniques have been developed that have resulted in identification, development, and release of FAW leaf-feeding resistant maize germplasm (Davis et al. 1989, Williams & Davis 1989, and Widstrom et al. 1993). Occasionally, results from our screening experiments have been poorer than expected or desired. When this happens, the research program suffers from loss of researcher(s) time, funds, seed, and progress. The researchers at Tifton have expressed concern over the virulence of their FAW culture since infusion of wild genes is not routinely done. Also, they have experienced poor establishment of FAW on both susceptible and resistant genotypes when the screening experiments were planted late in the growing season. Both research groups have observed that smaller plants are often more heavily damaged than larger plants of the same genotype. In 1993, the researchers at Mississippi State experienced a poor screening year with the FAW. Possible causes were insect colony, limited insect supply, and procedures used to process eggs and larvae for field infesting or some combination of these factors. Environment is also a possible factor, but is not under the control of the researcher(s). The present study was conducted to determine what effects insect colony, date of planting (early vs late), and plant growth stage when infested may have on screening maize for FAW leaf-feeding resistance.

MATERIALS AND METHODS

Field experiments were conducted at Mississippi State, MS and Tifton, GA during the 1995 growing season.

Insect Colony. At both Tifton and Mississippi State, screening for leaf-feeding resistance requires that each plant within the experiments be infested with approximately the same number of neonate FAW to provide a uniform infestation of the pest. Neonates for infesting were obtained from laboratory colonies. Eggs from the Mississippi colony were shipped by overnight mail to Tifton when plants there reached the appropriate growth stage and vice versa for Georgia colony eggs to Mississippi. Personnel at each location have developed their own procedures for laboratory rearing of FAW (Burton & Perkins 1989, Davis 1989). Their procedures differ primarily in larval diets, handling of adults and eggs, and genetic maintenance of the laboratory colonies. The Georgia colony has been maintained on a modified pinto bean diet for about six years in the laboratory without any infusion of wild genes (personal communication: W. D. Perkins), whereas, the Mississippi colony, which is reared on a wheatgerm-casein diet, has an infusion of wild genes almost every year.

Planting Dates. Researchers at both locations have utilized early and late plantings for screening. Sometimes inclement weather dictates later plantings and, on occasion early and a late plantings of the same maize genotypes are used so that experiments can be repeated within the same growing season.

In our experiments, there were two planting dates within each of the early and late planting periods. This was done so that two whorl stages of growth would be available at the time of infestation. Planting dates were as follows: early - Georgia (Apr. 5 and 19), Mississippi (Apr. 6 and 20), late - Georgia (May 2 and 10), Mississippi (May 3 and 17).

Plant Growth Stages. Entomologists and plant geneticists normally select a preferred growth stage to infest when screening for resistance. At Tifton, the researchers have selected the V_4 stage, whereas, at Mississippi State the preferred stage is from V_{68} (based on the system devised by Ritchie & Hanway [1982] for identifying stages of maize development). Sometimes, infestation is not possible at the desired plant stage for various reasons. For example, if insect supplies are inadequate when plants are ready, the plants will have grown to advanced stages before infestation can occur.

The infestations for the early and late planting periods were made when the plants of the second planting within each period reached the $V_{\rm s}$ stage at Mississippi State and the $V_{\rm s}$ stage at Tifton. Growth stage of the plants of the first planting within each planting period was recorded the day of infestation.

Maize Hybrids. Two susceptible and two resistant single cross maize hybrids were selected to study the above factors and their effects on identifying different levels of susceptibility. The susceptible hybrids were 'AB24E \times Mp305' and 'SC229 \times Tx601', and the resistant hybrids were 'Mp496 \times Mp708' and 'Mp704 \times Mp707'.

Experimental Design. The experiments were conducted using a randomized complete block design with treatments in a factorial arrangement with six replications. Treatments were 2 insect cultures, early and late plantings, 2 plant growth stages, and 4 single cross maize hybrids. The treatments were represented in each replication by a single plot of maize 6.1 m long and 0.9 m apart. Plots were thinned to approximately 20 plants before infestation. Accepted recommended agronomic practices were followed at both locations.

Infestation Technique. Each plant was infested with 30 neonate FAW mixed in 20/40 size corn cob grits and delivered into each whorl by a hand-held plastic device (Mihm 1983; Davis et al. 1989) when the plants of the second planting of each planting period (early or late) reached the specified growth stage. Plants of both plantings within each planting period were infested on the same day. The infestation dates were as follows: Mississippi State - early period plantings on May 25 and late period plantings on June 16; Tifton - early period plantings on May 12 and late period plantings on June 6.

Data. Each plant within a plot was visually scored for the degree of leaf-feeding damage on the 7th and 14th day after infestation. The rating systems used were the ones developed by Davis et al. (1992) specifically for the FAW. Both 7- and 14-day rating scales are based on the type and number of feeding lesions present on the leaves. The scores range from 0 to 9 with 0 indicating no damage and 9 indicating heavy damage. The two rating scales are highly correlated in their ability to separate resistant from susceptible genotypes. The 7-day scale minimizes the effect of leaf damage caused by migrating mid- instar FAW larvae, whereas, the 14-day scale gives the researcher a view of the total feeding damage caused by the larvae and provides an opportunity to determine if the plants show any late resistance response to the insect that would not be apparent at 7 days after infestation.

Statistical Analyses. Plot means were used in the analyses. Experiments were analyzed by location because of the differences in growth stage at the time of infestation. Data from the early and late planting periods were combined by the day (7 or 14) in which leaf-feeding damage scores were taken and analyzed by use of ANOVA (SAS 1989). Means were separated by least significant difference test at the 5% level of probability.

RESULTS

Mississippi State

An error was made when infesting the plants of the early planting period that resulted in only 3 of the 6 replications being used in the statistical analyses. Three factor interactions were significant for the analyses of both 7- and 14-day leaf-feeding damage rating scores. Therefore, inferences have been made utilizing nonmarginal means.

Insect Colony. Differences between FAW colonies for the degree of leaf-feeding damage were determined by comparing means within planting date periods, within growth stages infested, and within and among hybrids. Differences in 7 days rating scores between colonies occurred only within the susceptible hybrid 'Ab24E × Mp305' for the early planting period (Table 1). In this case, the Mississippi colony produced significantly more damage (at both leaf stages) than the Georgia colony. In the late planting period, significant differences in leaf damage scores occurred for 6 of the 8 comparisons between colonies within hybrids and growth stages. Each time, the Mississippi colony produced more damage than the Georgia colony. However, both colonies ranked the hybrids somewhat similarly when comparisons were made between cultures within growth stages and planting dates (Table 1). Similar results were obtained between colonies when 14-day rating scores were analyzed (Table 2).

Planting Date. The effect of planting date periods on screening results was determined by comparing means from 7- and 14-day ratings within growth stages at infestation, within insect colonies, and within and among hybrids. When comparisons were made between mean 7-day leaf-feeding damage scores of early and late planting periods (within the $\rm V_{\rm s}$ stage and within insect colonies and hybrids), only 1 of the 8 comparisons showed a significant difference (Table 1). However, when the same comparisons were made for plants infested in the $\rm V_{\rm 10}$ stage, 5 of the 8 comparisons were significantly different. The rating scores were higher for the early planting period. Comparisons among hybrids (between early and late planting periods, within growth stage and insect colony) revealed that the separation of susceptible from resistant hybrids was basically the same for both planting periods.

Table 1. Leaf-feeding damage rating scores taken 7 days after infestation of two stages of whorl maize plants [resistant (R) and susceptible (S)] with 30 neonate fall armyworms each at Mississippi State, MS ($\overline{x} \pm SD$).

Farly Planting Period Late Planting Factor Factor		Farly Planting Period	ting Period			Late Plant	Late Planting Period	
		Total Trans	and the			-	c	Ī
		Insect Colony	Colony			Insect	Insect Colony	
	M	MS	GA	A	M	MS	GA	A
		Growth Stage	Growth Stage at Infestation			Growth Stage	Growth Stage at Infestation	
Maize Hybrid	N _s	V_{10}	V	V_{10}	V	V_{10}	V _s	\mathbf{V}_{10}
$Ab24E \times Mp305 (S)$	8.2 ± 0.4	7.1 ± 0.4	7.2 ± 0.5	5.5 ± 0.8	7.8 ± 0.4	5.9 ± 0.4	6.6 ± 0.5	5.0 ± 0.5
$SC229 \times Tx601 (S)$	8.0 ± 0.4	6.2 ± 0.9	7.7 ± 0.8	5.6 ± 0.2	7.7 ± 0.2	4.7 ± 0.3	6.7 ± 0.8	4.0 ± 0.2
$Mp496 \times Mp708 \ (R)$	5.9 ± 0.1	4.5 ± 0.5	5.1 ± 0.1	3.9 ± 0.2	6.0 ± 1.0	3.7 ± 0.4	4.4 ± 0.9	3.5 ± 0.5
$Mp704 \times Mp707~(R)$	4.6 ± 0.1	2.9 ± 0.5	4.4 ± 0.5	2.9 ± 0.5	5.1 ± 0.6	2.3 ± 0.4	4.2 ± 0.3	1.8 ± 0.2
LSD (0.05) values:								
	1. For compa	1. For comparisons among means within early planting = 0.8	means within e	arly planting	= 0.8			
	2. For compa 3. For compa	2. For comparisons among means within rate planting $= 0.0$ 3. For comparisons among means between plantings $= 0.7$	neans between	ate pianting = 1 plantings = 0.	7.			

Table 2. Leaf-feeding damage rating scores taken 14 days after infestation of two stages of whorl maize plants [resistant (R) and susceptible (S)] with 30 neonate fall armyworms each at Mississippi State, MS $(\bar{x} \pm SD)$.

		Early Planting Period	ting Period			Late Planting Period	ing Period	
		Insect Colony	Colony			Insect Colony	Colony	
	M	MS	GA	A	M	MS	GA	A
		Growth Stage at Infestation	at Infestation			Growth Stage at Infestation	at Infestation	
Maize Hybrid	V _s	V_{10}	Vs	\mathbf{V}_{10}	V _s	V_{10}	V	V_{10}
$Ab24E \times Mp305 (S)$	8.2 ± 0.1	8.0 ± 0.2	7.2 ± 0.9	7.2 ± 0.3	8.9 ± 0.2	7.3 ± 0.6	8.1 ± 0.7	6.5 ± 0.7
$SC229 \times Tx601 (S)$	7.9 ± 0.1	7.8 ± 0.4	7.8 ± 0.2	8.0 ± 0.8	8.8 ± 0.3	6.2 ± 0.6	8.1 ± 0.4	5.2 ± 0.6
$Mp496 \times Mp708 \ (R)$	7.0 ± 0.1	6.5 ± 0.6	6.9 ± 0.3	5.8 ± 0.3	7.5 ± 0.4	5.5 ± 0.7	6.2 ± 1.1	4.5 ± 1.1
$Mp704 \times Mp707 (R)$	5.9 ± 0.8	5.5 ± 0.6	5.3 ± 0.8	4.8 ± 0.8	6.6 ± 0.6	4.0 ± 0.6	5.5 ± 0.4	4.6 ± 0.5
LSD (0.05) values:								
	1. For compa	1. For comparisons among means within early planting = 1.0	means within e	arly planting	= 1.0			
	2. For compa	2. For comparisons among means within late planting = 0.7	means within l	ate planting =	0.7			
	3. For compa	rrisons among 1	means between	$\mathfrak{a}_{\mathrm{n}}$ early and late	3. For comparisons among means between early and late plantings = 0.9	6		

The results of the 14 day ratings were similar to those obtained for the 7-day ratings (Table 2). Significant differences between early and late planting periods (within hybrids, insect colonies, and growth stages) occurred only when the plants were in the V_{10} leaf stage at infestation. Again, plants of the early planting period suffered more damage than those of the late planting period.

Plant Growth Stage. The effect of plant growth stage on screening results (damage rating scores) was determined by comparing means from 7- and 14-day ratings within planting date periods, within insect colonies, and within and among hybrids. Regardless of planting date period or insect colony, all comparisons between growth stages within hybrids were significantly different for the 7 day ratings (Table 1). In each case, the $\rm V_{10}$ plants sustained less damage than the $\rm V_{8}$ plants. Comparisons among hybrids within growth stage, planting date period, and insect colony revealed that resistant hybrids were separated from susceptible hybrids regardless of growth stage at infestation.

When the same comparisons were made for 14-day ratings (Table 2), only 1 of 8 comparisons were significant during the early planting period, whereas all comparisons in the late planting period showed significant differences between growth stages. Again, the V_{10} plants suffered less damage than the V_8 plants.

In general, the susceptible and resistant hybrids separated out as expected when comparisons were made between growth stages within insect colony and planting date period. However, the separation, especially of 'SC229 \times Tx601' and 'Mp496 \times Mp708', was not as clearcut as occurred for 7-day ratings.

Tifton

Interactions among factors were also significant at Tifton. There was a significant planting date period \times insect colony interaction for the 7-day rating scores and a significant planting date period \times hybrid \times insect colony interaction for the 14-day rating scores. Since these interactions were encountered, our inferences were made using nonmarginal means. The same comparisons as described for interpreting the Mississippi State data were used to study the effects that insect colony, planting date period, and plant growth stage may have on screening results.

Insect Colony. Only 3 of 16 comparisons between insect colonies (within planting date period and plant growth stage) were significant when the 7-day rating data were analyzed (Table 3). These significant comparisons were observed only in the early planting period and involved only the resistant hybrids. In each case, the Mississippi colony caused more damage than the Georgia colony. The hybrids were separated as to level of susceptibility similarly by either insect colony.

Results similar to those found for 7-day ratings occurred when the 14-day ratings were analyzed (Table 4). Significant differences between cultures occurred only in the early planting period and within the resistant hybrids. Again, the Mississippi colony caused more damage than the Georgia colony.

No differences were observed between insect colonies for separation of susceptible from resistant hybrids in the late planting period. However, a few differences between insect colonies did occur in the early planting period. For example, when plants were infested in the V_4 stage, there were no significant differences in rating scores among the hybrids infested with the Mississippi colony, whereas hybrids infested at the same growth stage with the Georgia colony separated out according to the expected resistant and susceptible categories.

Planting Date. Only 2 of 16 comparisons within insect colonies, plant growth stages, and hybrids showed significant differences between planting date periods when 7-day ratings were analyzed (Table 3). Both occurred within the resistant hy-

Table 3. Leaf-feeding damage rating scores taken 7 days after infestation of two stages of whorl maize plants [resistant (R) and stisceptible (S)] with 30 neonate fall armyworms each at Theon (A $(\overline{x} + SD)$

AIND SUSCEFIE	ыс (С) жин о	O NEONAIE FAL	AIND SUSCEPTIBLE (S)] WITH SO DECIMELY FALL ARMINOTHS EACH AT TIPTON, OF $(A \pm BD)$.	EACH AL LIFIC	in, GA (A ± SD)			
		Early Plan	Early Planting Period			Late Plan	Late Planting Period	
		Insect	Insect Colony			Insect	Insect Colony	
	2	MS	GA	A	Z	MS	GA	A
		Growth Stage	Growth Stage at Infestation			Growth Stage	Growth Stage at Infestation	
Maize Hybrid	V_4	V	V_4	V _s	V_4	V	V	V _s
$Ab24E \times Mp305 (S)$	9.0 ± 0.0	7.0 ± 0.6	8.8 ± 0.4	6.0 ± 1.1	8.0 ± 0.9	7.0 ± 2.1	8.2 ± 0.8	6.0 ± 2.4
$SC229 \times Tx601 (S)$	9.0 ± 0.0	7.0 ± 0.6	8.7 ± 0.5	6.0 ± 0.6	8.0 ± 0.6	6.3 ± 1.9	8.0 ± 0.8	6.5 ± 1.4
$Mp496 \times Mp708 \ (R)$	8.8 ± 0.4	6.3 ± 2.1	8.5 ± 0.5	3.7 ± 2.5	7.8 ± 0.8	2.8 ± 0.8	7.2 ± 0.8	3.7 ± 1.0
$Mp704 \times Mp707 \; (R)$	7.5 ± 1.0	3.0 ± 0.9	6.0 ± 1.7	1.7 ± 0.8	5.8 ± 1.0	2.3 ± 1.4	5.0 ± 0.7	1.7 ± 0.8
LSD (0.05) value = 1.3	1.3							

Table 4. Leaf-feeding damage rating scores taken 14 days after infestation of two stages of whore maize plants [resistant (R)

		Early Plan	Early Planting Period			Late Plan	Late Planting Period	
		Insect	Insect Colony			Insect	Insect Colony	
	N.	MS	GA	A	M	MS	GA	A
		Growth Stage	Growth Stage at Infestation			Growth Stage	Growth Stage at Infestation	
Maize Hybrid	V_{4}	V 8	V	N S	V	V	V_4	V ₈
$Ab24E \times Mp305 (S)$	9.0 ± 0.0	9.0 ± 0.0	9.0 ± 0.0	9.0 ± 0.0	9.0 ± 0.0	8.8 ± 0.4	9.0 ± 0.0	8.3 ± 0.8
$SC229 \times Tx601 (S)$	9.0 ± 0.0	9.0 ± 0.0	9.0 ± 0.0	9.0 ± 0.0	8.8 ± 0.4	9.0 ± 0.0	9.0 ± 0.0	8.8 ± 0.4
$Mp496 \times Mp708 \ (R)$	8.2 ± 0.4	8.7 ± 0.5	6.8 ± 1.7	7.5 ± 1.1	7.2 ± 0.8	6.3 ± 1.0	7.0 ± 1.3	7.0 ± 1.3
$Mp704 \times Mp707~(R)$	8.3 ± 0.8	6.5 ± 0.8	7.0 ± 1.5	5.7 ± 0.8	7.0 ± 0.6	3.8 ± 1.5	6.4 ± 1.1	3.8 ± 1.3
LSD (0.05) value = 0.9	6.0							

brids with lower rating scores being given to the plants of the late planting. Generally, the 7-day ratings from early and late planting periods resulted in no differences being observed as to separation of hybrids into resistant and susceptible categories.

Four of the 16 comparisons between planting date periods were significant when 14-day ratings were analyzed (Table 4). The differences all occurred within the resistant hybrids. As with the 7-day ratings, lower rating scores were attained in the late planting period plants. Planting date did not appreciably affect the separation of hybrids into susceptible and resistant categories (Table 4).

Plant Growth Stage. When comparisons were made between V_4 and V_8 growth stages within insect colonies, planting date periods, and hybrids (7-day ratings), all but one of the comparisons were significant (Table 3). The V_4 stage plants at infestation received higher rating scores than those infested at the V_8 stage. There were some differences that occurred at the 7-day rating as to separation of hybrids into resistant and susceptible categories. These differences involved the resistant hybrid 'Mp496× Mp708' when infested primarily at the V_4 stage. In these cases, no significant differences in rating scores were detected between this hybrid and the two susceptible hybrids.

Fewer differences between growth stages were detected when the 14-day ratings were analyzed (Table 4). The only significant differences were found within the resistant hybrid 'Mp704 \times Mp707'. This difference was significant regardless of planting date or insect colony. In each case, the $V_{\rm s}$ stage plants sustained less feeding damage. The separation of resistant from susceptible hybrids was not as clearcut when plants were infested at the $V_{\rm s}$ stage than when infested at the $V_{\rm s}$ stage.

DISCUSSION

Concern over the effect that continuous laboratory rearing of an insect on an artificial diet can have on field screening for resistance has been expressed by many plant resistance researchers. Guthrie et al. (1974) found that European corn borer, Ostrinia nubilalis (Hübner), laboratory colonies of up to one year in age (approximately 12 generations) could be used to successfully screen maize for resistance to this pest. However, European corn borer colonies maintained in the laboratory for longer periods of time were observed to begin losing their virulence. By the 46th laboratory generation, the borer larvae were found to cause little damage to field grown maize plants and were deemed to be no longer usable for screening purposes.

Quisenberry & Whitford (1988) observed differences in damage to bermudagrass cultivars depending upon the strain of FAW (bermudagrass/rice strain vs the maize strain) and the diet used to maintain the laboratory colonies. They suggested that FAW strain and laboratory diet be considerations when screening for bermudagrass resistance.

In our study, some significant differences in damage rating scores within hybrids were detected between FAW colonies. When this occurred, the Mississippi colony (with routine infusion of wild genes) was always found to cause more leaf damage than the Georgia colony. Both laboratory colonies were capable of causing sufficient damage so that resistant genotypes could be separated from susceptible genotypes. Our results did show that a FAW colony can be maintained in the laboratory on an artificial diet for at least 6 years (approximately 72 generations) without losing its ability to cause sufficient leaf damage to separate maize genotypes appropriately as to susceptibility level. However, the Georgia colony appears to have lost some of its virulence when compared to the Mississippi colony.

Also, our results showed that planting date (early vs late) was not a factor at either location in affecting the successful separation of resistant from susceptible genotypes.

Therefore, a researcher could expect similar results if a screening experiment was repeated in the same year.

Williams et al. (1983) suggested that resistance mechanisms operating in FAW resistant maize plants may have been more strongly expressed at the 10-leaf stage than the 5-leaf stage. Videla et al. (1992) reported that FAW resistant maize was resistant to this pest throughout the vegetative growth stages based on larval growth and survival. However, no consistent differences between larval growth or survival were found among plant growth stages within the test single cross maize hybrids. Wiseman & Isenhour (1993) observed that commercial maize hybrids in the 12-leaf stage, when infested with neonate FAW, tolerated damage by this pest better than plants at the 8-leaf stage at infestation.

Our results, especially the 7-day ratings at both locations (Tables 1 and 3), show that FAW feeding damage occurred less on the older than on the younger plants for both resistant and susceptible hybrids. In general, this difference in growth stage susceptibility did not alter the separation of resistant from susceptible hybrids. However, the best separations among hybrids seemed to occur when the plants were $V_{\rm s}$ stage when infested. Researchers should be aware that rating scores within experiments could reflect differences in plant stage because of dissimilar growth rates among genotypes. Also, plants of different sizes within a plot caused by variation in germination can result in a somewhat inaccurate rating score. When thinning or infesting plants within a plot, plants not in the appropriate stage should be eliminated. These data on differences in rating scores between plant growth stages confirm the suggestion by Williams et al. (1983) concerning the increased expression of resistant mechanisms in $V_{\rm s}$ and older stage plants. Future research on elucidating the factors responsible for this resistance should consider comparing magnitude of factor expression in plants of different growth stages.

In conclusion, significant differences in rating scores within each factor (insect colony, planting date period, and plant growth stage) were found for some comparisons. However, none of these factors (at least in this study) were found to appreciably alter the separation of resistant from susceptible genotypes, which is the objective of screening.

When a screening failure occurs, it is important to determine, if possible, the factor or factors responsible. Under other circumstances, the three factors studied here may play a role. Since these data do not present a clear picture on the effect of insect colonization on the ability of resulting FAW larvae to damage maize plants, we suggest a regular infusion of wild genes into the laboratory colonies. Other factors should also be investigated, such as breakdowns in the proper procedures for handling eggs and neonate larvae for field infesting. We consider one of the best ways to avoid screening failures is to have an ample supply of healthy, highly virulent larvae at the time the plants reach the desired growth stage. Further, it is recommended that plants be infested at or near the \mathbf{V}_{s} stage for the best separation of test genotypes for leaf-feeding damage.

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