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## GENETIC ANALYSIS OF RESISTANCE TO EUROPEAN CORN BORER (*OSTRINIA NUBILALIS* HUB. LEPIDOPTERA:CRAMBIDAE) DAMAGE IN EIGHT MAIZE GERMPLASM

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### Abstract

The inheritance of resistance to *Ostrinia nubilalis* (ECB) damage in eight breeding maize lines was studied by Griffing's diallel analysis (Griffing 1956) under two water regimes. One hundred fifty neonate larvae of second generation of ECB were infested around ear node of maize germplasm and approximately two months later, the potentials of the germplasm were determined by measuring stalk and shank tunneling damage in plants. General combining ability (GCA) was more important than specific combining ability (SCA) in determining resistance to both stalk and shank tunneling. Although mean squares for GCA accounted for 76.1% of the variation for stalk tunneling and 70.6% of the variation for shank tunneling, the performance of some crosses between resistant and susceptible lines indicated some dominant genes may have role in resistance mechanism. In general, the results suggest that resistance may be improved with recurrent selection methods within this germplasm.

**Keywords:** *Ostrinia nubilalis*, resistance, breeding, diallel analysis

### Sekiz Farklı Mısır Hattında Mısır Kurdu (*Ostrinia nubilalis* Hub. Lepidoptera:Crambidae) Zararına Karşı Dayanıklılık Mekanizmasının Genetik Analizi

### Özet

Bu çalışmada iki farklı sulama rejiminde yetiştirilen 8 mısır hattında *Ostrinia nubilalis* (Mısır Kurdu)'a karşı dayanıklılık mekanizması Griffing'in diallel analizi (Griffing 1956) yöntemine dayalı olarak araştırılmıştır. Bu amaçla, ikinci generasyon mısır kurdu ilk dönem larvaları, bitkilerin anthesis döneminde koçan nodu bölgesinde 150 adet olarak birer hafta arayla aşılansın ve takriben 2 ay sonra bitkiler, gövde ve koçan sapında meydana gelen tüneller ölçülerek hatların dayanıklılık bakımından genetik potansiyelleri belirlenmiştir. Gövde ve koçan sapındaki zararlanmalara karşı dayanıklılık verileri, GCA'nın (General Combining Ability) SCA'dan (Specific Combining Ability) daha önemli olduğunu ortaya koymuştur. Gövde tünellerine ait varyasyonun %76.1 ve koçan sapına ait varyasyonun %70.6 sını GCA oluşturmalarına rağmen, bazı dayanıklı ve hassas hatlar arasında oluşturulan melezlerin gösterdiği performans, dominant genlerinde dayanıklılık mekanizmasında rol oynayabileceğini göstermiştir. Genel olarak sonuçlar, elde bulunan mısır hatları ile dayanıklılığın tekrarlamalı seleksiyon ile geliştirilebileceğini göstermektedir.

**Anahtar kelimeler;** *Ostrinia nubilalis*, Dayanıklılık, Islah, Diallel analizi

### 1. Introduction

Maize, *Zea mays* L., is the third largest most important cereal crop of the world after rice and wheat (FAO 1974). The European corn borer (ECB), *Ostrinia nubilalis*, (Hubner)(Lepidoptera:Crambidae) is considered to be a major pest insect of maize, and distributed throughout the Middle East, North Africa, Europe and North America. The use of resistant plants to reduce crop losses caused by insects is an effective, and economically and environmentally acceptable method of pest control. Therefore, development of resistant maize lines to European corn borer larval

feeding has been a cornerstone of maize breeding programs (Barry and Darrah 1991).

The success of any breeding programs depends on understanding of the genetics of characters. Several methods have been developed for investigating the components of genetic variance. Sprague and Tatum (1941) were the first to introduce the concept of general combining ability (GCA) and specific combining ability (SCA) to the genetic studies. They defined GCA as the average performance of a line in hybrid combinations and SCA as the performance of specific crosses in which hybrid

combinations are either better or poorer than expected based on the average performance of the parents lines included. Among those genetic analysis, the diallel mating design has been used more extensively than any other mating design in maize and other crop species (Hallauer and Miranda 1988) since it was first proposed by Yates (1947). Therefore, the objective of the current study was to obtain information on the gene action governing second generation European corn borer resistance in eight breeding maize lines and 28 F<sub>1</sub> single crosses in a diallel mating design.

## 2. Materials and Methods

A 2-year field experiment was conducted under two water regimes, irrigated and non-irrigated, on a Kennebec silt loam soil at the University of Nebraska, Department of Agronomy Research Farm, Lincoln, NE during the 1996 and 1997 growing seasons. Eight S<sub>1</sub> maize lines, previously selected for ECB resistance were used as parents in this study. Eight parents, 28 F<sub>1</sub> crosses and four commercial checks were machine planted in a randomized incomplete block design with two replications in both years. Thirty and twenty-five kernels per row were planted and later thinned to 25 and 20 kernels resulting in a final plant population of 54,500 and 43,700 plants/ ha in the irrigated and non-irrigated water regimes, respectively.

To simulate natural infestation, the plants in one of the rows in each plot were manually infested with neonate ECB larvae (from French Agricultural Research, Lamberton, MN) to ensure uniform ECB infestation. A "bazooka" designed by Mihm et al. (1978) was used to infest plants at the ear node, one node above and one node below the ear node. These sites were chosen because second generation moths prefer to lay their eggs around the ear node. Each node was infested two times with 50 neonate larvae per node during anthesis of the earliest and latest entries. Evaluation of all entries for resistance to second generation

damage began approximately 50-60 days after the second manual infestation. Five manually and five naturally infested plants from each entry were examined for stalk and shank tunneling injury. Stalk injury was determined by splitting the stalk and counting the number of cavities per plant. Each 2.54 cm. of stalk tunneling was considered to be equal to one cavity. Shank damage ratings were based on whether there was tunneling damage in the main ear shank and thus, each plant was ranked as 0 or 1. Shank ratings was transformed by using an arcsin transformation before statistical analysis to better interpret the data (Steel et al. 1997).

The data were initially analyzed including commercial hybrid controls, parental inbreds and crosses to test the null hypothesis that there were no differences among entries. Analysis of variance (SAS Procedure Mixed) was performed for all data. After completion of the initial analysis, the data were reanalyzed without commercial checks to access unbiased estimates of parents and cross means, and their corresponding error terms for use in the diallel analyses. Diallel analyses for stalk and shank tunneling resistance were based on the performance of entries over water regimes averaged in two years. A fixed effect model was assumed because parents were not randomly chosen. Therefore, Griffing's experimental Method 2, Model 1 analysis was performed where variation among crosses and parents was partitioned into components for general combining ability (GCA) and specific combining ability (SCA) (Griffing 1956).

## 3. Results

Relative genotype reaction was similar in both years for water regimes and infestation levels. Therefore, the data for both damage ratings were combined. For stalk tunneling, seven out of eight parents and all of the crosses were found to be as resistant or more resistant than the most resistant commercial check Mycogen 7250CB (Table1). In fact, twelve of 28

Table 1. Combined resistance means of maize inbreds, crosses and commercial checks for second generation european corn borer stalk and shank damage.

Entry	Inbreds and Crosses	Stalk Rating Means	Shank Rating Means*	Entry	Inbreds and Crosses	Stalk Rating Means	Shank Rating Means*
1	P <sub>1</sub> = (NECB 5) -2-3	3.18	0.69	21	P <sub>2</sub> P <sub>8</sub>	2.82	0.87
2	P <sub>2</sub> = (NECB 7) -1-1	3.72	0.88	22	P <sub>3</sub> P <sub>4</sub>	3.19	0.89
3	P <sub>3</sub> = (NECB 9) -6-3	6.29	1.20	23	P <sub>3</sub> P <sub>5</sub>	2.75	0.79
4	P <sub>4</sub> = (NECB 14) -3-1	3.16	1.07	24	P <sub>3</sub> P <sub>6</sub>	3.82	0.93
5	P <sub>5</sub> = (NECB 15) -9-2	3.57	1.00	25	P <sub>3</sub> P <sub>7</sub>	3.19	1.14
6	P <sub>6</sub> = (NECB 16) -9-2	3.72	0.88	26	P <sub>3</sub> P <sub>8</sub>	3.25	0.81
7	P <sub>7</sub> = (NECB 18) #EXP-3	3.19	0.88	27	P <sub>4</sub> P <sub>5</sub>	2.12	0.61
8	P <sub>8</sub> = (NECB 20) -1-2	3.83	1.24	28	P <sub>4</sub> P <sub>6</sub>	2.12	0.63
9	P <sub>1</sub> P <sub>2</sub>	2.10	0.66	29	P <sub>4</sub> P <sub>7</sub>	2.03	0.66
10	P <sub>1</sub> P <sub>3</sub>	2.81	0.76	30	P <sub>4</sub> P <sub>8</sub>	2.05	0.70
11	P <sub>1</sub> P <sub>4</sub>	1.80	0.69	31	P <sub>5</sub> P <sub>6</sub>	2.44	0.83
12	P <sub>1</sub> P <sub>5</sub>	1.91	0.76	32	P <sub>5</sub> P <sub>7</sub>	2.82	0.83
13	P <sub>1</sub> P <sub>6</sub>	2.11	0.65	33	P <sub>5</sub> P <sub>8</sub>	3.38	0.90
14	P <sub>1</sub> P <sub>7</sub>	2.15	0.61	34	P <sub>6</sub> P <sub>7</sub>	2.93	0.66
15	P <sub>1</sub> P <sub>8</sub>	2.21	0.60	35	P <sub>6</sub> P <sub>8</sub>	3.50	1.02
16	P <sub>2</sub> P <sub>3</sub>	3.17	0.91	36	P <sub>7</sub> P <sub>8</sub>	2.63	0.78
17	P <sub>2</sub> P <sub>4</sub>	1.58	0.60	37	ASGROW RX 801	3.69	1.11
18	P <sub>2</sub> P <sub>5</sub>	2.67	0.76	38	PIONEER 3225	3.54	1.01
19	P <sub>2</sub> P <sub>6</sub>	2.16	0.65	39	MYCOGEN 7250CB	3.36	0.99
20	P <sub>2</sub> P <sub>7</sub>	3.03	0.85	40	HOEGEMEYER 2626	3.91	0.77

Standard error of entry differences 0.48 0.17

\*Arcsine transformed data means

single crosses were more resistant than the most resistant check for stalk tunneling.

The stalk damage rating of the best single cross (P<sub>2</sub> x P<sub>4</sub>) was more than two times greater than that of the best check. Although none of the parental lines or single crosses showed better shank resistance than the most resistant commercial check (Hoegemeyer 2626), one of eight parental lines (P<sub>1</sub>) and 15 of 28 single crosses had numerically lower resistance ratings than that of Hoegemeyer 2626.

The data for all measured traits were reanalyzed without commercial checks in order to obtain unbiased estimates of combining abilities of the single crosses and their corresponding parents. For this study, general combining ability (GCA) effects were important for both resistance traits studied while specific combining ability (SCA) effects were not significant for shank tunneling ratings. Mean squares for GCA accounted for 76.1% of the variation for

stalk tunneling, and 70.6% for shank tunneling (Table 2). The breeding values of the S<sub>1</sub> lines for each resistance trait were evaluated by estimating their GCA effects. For stalk tunneling, the parents P<sub>1</sub>, P<sub>3</sub>, and P<sub>4</sub> had GCA effects that differed from zero (Table 3). P<sub>1</sub> and P<sub>4</sub> were the most resistant parents with negative GCA effects of -0.430, and -0.464, respectively, whereas the most susceptible parent was P<sub>3</sub> with the highest positive GCA effects of 0.891. The remaining parents showed intermediate GCA effects that did not differ from zero. The estimates of GCA effects for shank tunneling suggested that P<sub>1</sub> (-0.123) was the best source for this trait and it also had the second best GCA value for stalk tunneling (Table 3). Conversely, as it was for stalk tunneling, P<sub>3</sub> was the most susceptible parent for shank tunneling with a positive GCA value of 0.130 (Table 3). The inbreds of P<sub>2</sub> and P<sub>4</sub> also contributed shank resistance to their progenies as indicated by

Table 2. Mean squares of diallel set of eight parents and their 28 single crosses for stalk and shank tunneling ratings.

Source of variation	df	Stalk tunneling	Shank tunneling
GCA	7	1.78*	0.060*
SCA	28	0.56*	0.025
Error	34	0.31	0.034
Percentage of Mean square for GCA		76.1	70.6
Percentages of Mean square for SCA		23.9	29.4

\*: Significant at the 0.05 probability levels

Table 3. Estimates of GCA effects of eight parental lines for stalk and shank tunneling ratings.

Parents	General combining ability estimate	
	Stalk tunneling	Shank tunneling
P <sub>1</sub>	-0.430*	-0.123
P <sub>2</sub>	-0.086	-0.029
P <sub>3</sub>	0.891*	0.130*
P <sub>4</sub>	-0.464*	-0.042
P <sub>5</sub>	-0.062	0.011
P <sub>6</sub>	0.066	-0.022
P <sub>7</sub>	-0.071	-0.0069
P <sub>8</sub>	0.160	0.081
s.e. (g <sub>i</sub> ) <sup>1</sup>	0.1492	0.0565
s.e. (g <sub>j</sub> -g <sub>i</sub> ) <sup>2</sup>	0.2375	0.1456

\*: Significant at the 0.05 probability level.

<sup>1</sup>= standard error of GCA effect.

<sup>2</sup>= standard error of the differences between GCA effects.

the GCA effects of -0.029 and -0.042, respectively. However, the effects were not as large as that of P<sub>1</sub>. Furthermore, none of the parents showed significant mixed performance with respect to resistance to both stalk and shank tunneling. P<sub>1</sub> and P<sub>4</sub> always contributed stalk and shank resistance to their progenies whereas the parent P<sub>3</sub> always appeared to be most susceptible parent with respect to GCA effects of stalk and shank tunneling damage (Table 3). This was indication of some common genes for resistance to both type of damage.

Specific combining ability effects varied greatly among the 28 crosses for stalk tunneling whereas SCA was not statistically significant for shank tunneling. The range of SCA effects for stalk damage was from

-0.948 for the cross P<sub>3</sub>xP<sub>5</sub> to 0.40 for P<sub>5</sub>xP<sub>7</sub> (Table 4). With respect to stalk tunneling,

Table 4. Estimates of SCA effects for 28 single crosses for stalk tunneling ratings.

Cross	Specific Combining Ability Estimate		
	SCA	Cross	SCA
P <sub>1</sub> P <sub>2</sub>	-0.242	P <sub>3</sub> P <sub>5</sub>	-0.948*
P <sub>1</sub> P <sub>3</sub>	-0.515	P <sub>3</sub> P <sub>6</sub>	-0.009
P <sub>1</sub> P <sub>4</sub>	-0.168	P <sub>3</sub> P <sub>7</sub>	-0.504
P <sub>1</sub> P <sub>5</sub>	-0.463	P <sub>3</sub> P <sub>8</sub>	-0.680*
P <sub>1</sub> P <sub>6</sub>	-0.382	P <sub>4</sub> P <sub>5</sub>	-0.228
P <sub>1</sub> P <sub>7</sub>	-0.216	P <sub>4</sub> P <sub>6</sub>	-0.356
P <sub>1</sub> P <sub>8</sub>	-0.391	P <sub>4</sub> P <sub>7</sub>	-0.308
P <sub>2</sub> P <sub>3</sub>	-0.506	P <sub>4</sub> P <sub>8</sub>	-0.528
P <sub>2</sub> P <sub>4</sub>	-0.745*	P <sub>5</sub> P <sub>6</sub>	-0.436
P <sub>2</sub> P <sub>5</sub>	-0.048	P <sub>5</sub> P <sub>7</sub>	0.076
P <sub>2</sub> P <sub>6</sub>	-0.696*	P <sub>5</sub> P <sub>8</sub>	0.406
P <sub>2</sub> P <sub>7</sub>	-0.319	P <sub>6</sub> P <sub>7</sub>	0.058
P <sub>2</sub> P <sub>8</sub>	-0.126	P <sub>6</sub> P <sub>8</sub>	0.397
P <sub>3</sub> P <sub>4</sub>	0.106	P <sub>7</sub> P <sub>8</sub>	-0.338
s.e.(s <sub>ij</sub> ) <sup>1</sup>	0.348		
s.e.(s <sub>ij</sub> ) <sup>2</sup>	0.697		
s <sub>ik</sub> <sup>2</sup>			

\*: Significant at the 0.05 level.

<sup>1</sup>= standard error of SCA effect.

<sup>2</sup>=standard error of the difference between SCA effects.

five out of 10 lowest SCA effects involved the most susceptible inbred P<sub>3</sub>, whereas four out of 10 lowest SCA effects crosses involved at least one parent with low GCA effect (P<sub>1</sub> or P<sub>4</sub>). The best hybrid combination (P<sub>1</sub> x P<sub>4</sub>) showed the second best SCA performance for stalk tunneling.

#### 4. Discussion

The levels of resistance of these germplasm used in this study were moderately or mostly resistant when compared to commercial checks with the exception of the parental lines P<sub>3</sub> for stalk tunneling and P<sub>3</sub> and P<sub>8</sub> for shank tunneling. Genotypic variation was found for all

measured traits. GCA was the predominant factor explaining genetic variation among crosses. Even when SCA effects were found, GCA mean squares were at least three times larger than the SCA mean squares (Table 2). This implies the importance of additive gene action in the inheritance of resistance traits measured. These observations also confirm those of Onukogu et al. (1978); Kim et al. (1989); Lamb et al. (1994) who reported the predominant role of additive gene effects for resistance to second generation ECB damage as measured by sheath-collar ratings. The inbreds of  $P_1$  and  $P_4$ , exhibited the highest negative estimates of GCA effects for stalk tunneling (Table 3) indicating that these parental lines could contribute high resistance to their progenies. Similarly, the same parental lines showed high resistance for ECB shank tunneling. This relates that selecting for resistance to one type of ECB damage could increase the level of resistance to the other type of damage. This also suggests that at least some genes for resistance to stalk tunneling might confer resistance to shank tunneling.

## 5. Conclusion

This study confirms that both resistance traits evaluated in the eight maize parental lines are inherited on a quantitative basis. Although GCAs indicated the importance of additive gene action for ECB resistance, the results from crosses with resistant lines and susceptible line  $P_3$  indicated a dominant gene effect for stalk tunneling resistance. However, overriding importance of the additive component of variance suggests that active selection for

the improvement of the traits studied in this material should be based on recurrent selection methods that take advantage of additive type of gene action.

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