

LABORATORY ASSESSMENT OF THE INHERENT SUSCEPTIBILITY OF SOME MAIZE VARIETIES TO POST-HARVEST INSECT INFESTATION

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ABSTRACT

Four Egyptian maize cultivars: Sweet grain sorghum, three way cross 321, Giza 2 and Single cross 10-planted in two different dates were tested for their susceptibility to post harvest insect infestation. Two insect genera were only detected, *Sitotroga cerealella* Oliv. (Lepidoptera: Gelechiidae) and *Sitophilus zeamais* Motsch. (Coleoptera : Curculionidae)

The variety Giza 2 was the least cultivar attacked by insects considering the total number of emerged adults of both insect species. Both, difference in weight loss of infested grains and percentage of grain viability, in the four cultivars considered, were found significant. Based on the susceptibility indices (SI), the four cultivars were ascendingly arranged as Giza 2, Single cross 10, Sweet grain sorghum and the more susceptible one Three way cross 321. SDS-protein electrophoresis analysis proved that each cultivar had an unique band (UB) which could be used as a biochemical marker to discriminate among them.

Keywords: *S. cerealella*, *S. zeamais*, susceptibility indices, maize cultivars .

INTRODUCTION

Maize (*Zea mays* L.) is one of the most important cereal crops in Egypt. Host plant resistance to insects is environmentally safe, economically feasible and socially acceptable as a tactic of integrated pest management (IPM). Susceptible varieties of maize have been reported to suffer 30-40% loss due to attack by field pests (Kumar and Miham, 1995, 1996). The incorporation of genes for resistance in such susceptible germplasm has been reported to reduce the losses to less than 5%. The post harvest losses in maize caused by storage pests are enormous although authentic assessments are rare.

Maize, in storage was found to be attacked by several important cosmopolitan pests causing losses varied from 9 to 45% depending upon the period of storage (Pantenus, 1988; Markham *et al* 1991 and Kumar, 2002). The main objective of this work is to test a number of Egyptian maize lines for their resistance, evaluate their susceptibility indices to post harvest insect infestation and compare the protein patterns of the tested grain varieties.

MATERIALS AND METHODS

Estimation of damage caused by insect infestation:

Four Egyptian cultivars of maize grains were harvested from the experimental fields, Faculty of Agriculture, Ain Shams University at Shalakan, Qalybia Governorate. The maize cultivars were: Sweet grain sorghum, three

way cross 321, Giza 2 and Single cross 10, collected in two planting dates (24th April and 28th May) and then were stored under laboratory conditions (28±2°C and 70±5% RH) till adult emergence of the natural field infestation of the grains before storage.

From each variety, several replicates of 250 grams maize grains were weighted and placed in 500 cc clean glass jars. After one month and four months of storage, three replicates from each variety were externally examined for exit holes counting the emerging adult species, and the grains were sieved to remove dust and frass and then weighted. The percentage loss in grain weight was determined as the difference between initial and final weight using the following equation:

$$\% \text{ weight loss} = (IW - FW) \times 100 / IW$$

where IW is the initial weight and FW is the final weight of tested grains.

Germination tests were carried out according to International Standard Methods (ISM), (Anonymus, 1966). For this purpose, one hundred maize grains from each cultivar were placed in plastic trays divided into 100 sections (5 x 5 cm) containing continually moistened sandy soil for about two weeks then the number of germinated grains were recorded. The experiments were replicated three times.

Susceptibility of maize cultivars to Insect Infestation:

Ten grams from each of the four tested cultivars were weighed and placed separately into 100 ml glass jars. Three pairs of newly emerged adults of either *S. cerealella* or *S. zeamais* were placed in each jar for four days for mating and oviposition. Afterwards all adult insects were removed leaving the grains and deposited eggs in the jars till adult emergence of F₁ progeny which were counted. The developmental period (in days) of each insect on each cultivar was also recorded. Susceptibility indices (SI) were calculated according to Dobie (1974):

$$SI = \frac{\text{Log } F_1}{D} \times 100$$

Where: F₁, is the total number of emerging adults D, median developmental period (estimated as the time from the middle period of oviposition to the adult emergence of 50% of the number of emerging adults. The experiment was replicated three times.

Determination of protein fractions

Protein fractions according to their molecular weight was performed using polyacrylamide gel electrophoresis (PAGE) in the presence of sodium dodecyl sulphate (SDS) as described by Laemmli (1970) and modified by Studier (1973) using an acrylamide gradient (7% to 16%) gel. Gel was photographed and scanned with Bio-Rad video Densitometer model 620 and IBM compatible personal computer 165-2072 at a wavelength of 577. Electrophoresis in polyacrylamide gel has proven to be more successful tool for the separation of protein subunits and determination of their molecular weight.

Statistical analysis

All data collected were subjected to statistical analysis without transformation using two way analysis of variance (ANOVA). The means were separated using the least significant Difference test (LSD).

RESULTS AND DISCUSSION

Effect of maize cultivars on insect infestation:

Insect infestation in maize grains in the two planting dates and stored from one and four months are shown in Table (1). Two insect genera were only detected; *Sitotroga cerealella* Oliv. and *Sitophilus zeamais* Motschulsky. Grains planted in April and stored for one month, the variety Giza 2 was the least infested as the total number of emerged adults of both *S. cerealella* and *S. zeamais* was 0.8, while it was 40.8, 10.4 and 4.0 for Three way cross 321, Sweet grain sorghum and Single cross 10, respectively. It is worthwhile to mention that three way cross 321 was the most favourable variety to Lepidopterans than Coleopterans while it is vice versa with the other varieties. At the end of May, it was also found that variety Giza 2 was the least to be attacked (Nil) by both insects while Sweet grain sorghum was the most favourable one. Moreover, Coleopterans were more than Lepidopterans in all cases Table (1).

After storage for four months, Giza 2 was also the least cultivar attacked by insects as the mean number of infestation by Lepidopteran and Coleopteran insects were 0.4 and 10.4, respectively (total 10.8). The other grain cultivars could be arranged in the following ascending order in regard to total insect infestation, Sweet grain sorghum (39.3), Single cross 10 (58.3) and Three way cross 321 (88.6), (Table 1).

As a result of insect infestation, the weight of grains was affected. The percentage of weight loss of Giza 2 grains, less infested, was only reduced to 3.25 and 7.13 in April and May plantation, respectively. This result indicated that Giza 2 cultivar was the most resistant one for insect infestation. There was a good correlation between grain weight loss and number of emerged insects. Meanwhile, the losses ranged from 11.77% to 20.44% and 10.04% to 23.45% in both April and May plantation, respectively, for the other cultivars. These results demonstrated extensive feeding and reproduction of the insects on the susceptible cultivars. Kossou *et al* (1993) and Vowolor *et al* (1995) suggested that maize variety had a significant effect on egg incubation, mean duration, mean weights of most developmental stages of *S. zeamais* and the site of weevil emergence from the kernel. Barney *et al* (1991) and De and Sarup (1991) reported that resistance of stored maize varieties to *S. zeamais* was believed to be related to the chemical composition of the grains.

The difference in weight loss of grains due to insect infestation was significant in the four cultivars considered. The percentage of grain viability (detected as percent germination) was also significant. In both dates of cultivation, Giza 2 gave the highest viability, because, those grains did not provide an optimum niche for insect to feed and produce as freely as on the other tested cultivars.

Table 1: Percentage germination, weight of 1000 grains, number of *Sitotroga cerealella* and *Sitophilus zeamais* insects obtained from four maize varieties, planted in two dates.

Planting date	Cultivars	Weight of 1000 grains	% Germination	Sample weight (gm)	Insect Infestation									
					After one month			After four month of storage						
					Mean No. of l/pl.	Mean No. of Coleo.	Total	Mean No. of Lepi	Mean No. of Coleo	Total	% Infestation	Grains weight	% weight loss	% Germination
1 st 24/4/2001	Sweet grain sorghum	27.30	72	250	4.0	6.4	10.4	3.9	35.4	39.3	84.2	199.15	20.44	14
	Three cross 321	341.87	79	250	32.8	8.0	40.8	56.4	32.2	88.6	58.8	201.50	19.14	27
	Giza 2	333.16	90	250	0.0	0.8	0.8	0.4	10.4	10.8	9.4	241.87	3.25	69
	Single cross 10	347.47	68	250	1.8	2.2	4.0	10.5	47.8	58.3	66.8	220.57	11.77	41
2 nd 28/5/2001	Sweet grain sorghum	16.53	79	250	2.2	5.4	7.6	1.9	7.8	9.7	81.3	207.65	16.94	19
	Three cross 321	363.97	75	250	0.8	5.0	5.8	0.5	82.8	83.3	84.4	191.38	23.45	20
	Giza 2	310.13	86	250	0.0	0.0	0.0	16.3	9.9	26.2	24.2	232.17	7.13	34
	Single cross 10	357.23	86	250	0.4	1.0	1.4	24.6	1.7	26.3	72.6	224.90	10.04	33

Number of Insects emerged after four months

1st planting date 24 April

"F" between *S.cerealella* = 6.5647 (Significant) LSD = 31.3199

"F" between *S.zeamais* = 14.5420 (significant) LSD = 12.5740

2nd planting date 28 May

"F" between *S.cerealella* = 8.0500 (significant) LSD = 12.5866

"F" betwent*S.zeamais* = 14.6770(significant) LSD = 30.7900

Susceptibility of some maize cultivar to insect Infestation:

Data in Table (2) indicated that Giza 2, in the planting date late April, had a small number of emerged adults of both *S. cerealella* and *S. zeamais* (5.33 and 16.67). This was closely followed by Single cross 10 (12.67 and 34.67), and Sweet grain sorghum (30.0 and 30.0). Meanwhile, Three way cross 321 had the highest number of emerged adults (58.0 and 81.67). Based on the susceptibility indices (SI) the four cultivars could be arranged as follow: (i) slightly susceptible, Giza 2 and Single cross 10, (ii) moderately susceptible, Sweet grain sorghum and (iii) susceptible, Three way cross 321.

As for May planting date, susceptibility indices did not follow the same trend. The SI values of cultivars in this period indicated that they were generally less susceptible to insect infestation (in addition to their variable susceptibility to insect species infestation). It is noteworthy that the planting date 24th May was less suitable for insect emergence. For *S. cerealella* the SI indicated that the cultivars were ascendingly arranged as, Giza 2, Single cross 10, Sweet grain sorghum and the more susceptible one Three way cross 321, but for *S. zeamais* they were Giza 2, Single cross 10, Sweet grain sorghum and the highest Three way cross 321. From the above mentioned data, it is observed that lower values of SI indicate more resistance of grains to insect infestation. The results of the present studies, could provide useful information for the development of a pest management programme for these insects.

Table 2: Relative susceptibility of four maize cultivars to *Sitotroga cerealella* and *Sitophilus zeamais* infestation.

Cultivar	Mean No. of emerged adults		Mean developmental time (days)		Susceptibility Indices (SI)	
	S.	S.	S.	S.	S.	S.
	<i>cerealella</i>	<i>zeamais</i>	<i>cerealella</i>	<i>zeamais</i>	<i>cerealella</i>	<i>zeamais</i>
Sweet grain sorgh	30.0	30.0	31.0	25.33	4.76	5.83
Three way cross 321	58.0	81.67	29.0	25.0	6.08	7.65
Giza 2	5.33	16.67	30.0	29.0	2.42	4.21
Single cross 10	12.67	34.67	31.33	35.33	3.52	4.36
Sweet grain sorgh.	29.21	29.70	34.00	26.27	4.31	5.24
Three way cross 321	41.60	74.52	43.17	26.31	3.75	6.97
Giza 2	6.12	12.87	34.81	28.88	2.26	3.02
Single cross 10	10.97	10.31	35.99	32.94	2.89	2.11

SDS-protein electrophoresis

The buffer soluble proteins extracted from grains of the four tested cultivars were analyzed by sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE). As shown in Fig. (1) a total of 36 bands were characterized in the four maize cultivars. These bands had different relative mobilities (R_m), ranging from 0.055 to 0.643, also molecular weights (MW) ranging from 270.0 to 25.25 KDa. Each cultivar had an unique band (s) (UB) which could be used as a specific biochemical marker. The tolerant cultivar Giza 2 showed lower number of bands (around 12 bands) while it had a UB with R_m of 0.251 and a MW of 142.20 KDa which was not found in any of the other cultivars. The sensitive cultivar Sweet grain sorghum showed an absence of two bands with MW of 270.46 and 265.67 KDa and presence of

two bands with MW of 39.44 and 27.54 KDa. These results are in agreement with (Abdel-Tawab *et al* 2001 and 2002).

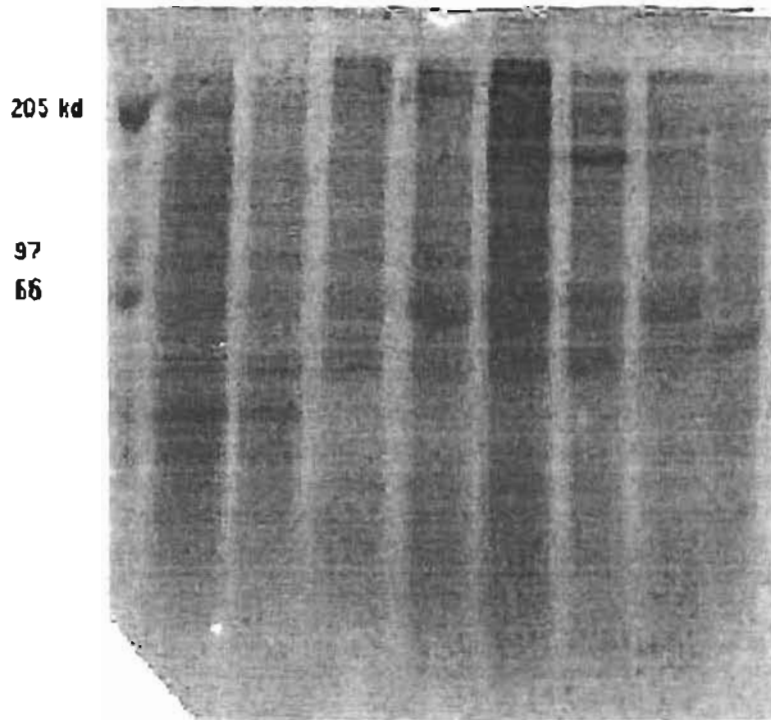


Fig. (1.). SDS-PAGE profiles of total proteins of the four tested maize cultivars planted in two different dates arranged from left to right:

1, Standard Marker

2,3 Sweet grain sorghum 1st and 2nd planting date

4,5 Three way cross 321 1st and 2nd planting date

6,7 Giza 2 1st and 2nd planting date

8, 9 single cross 10 1st and 2nd planting date

Generally, these observations would indicate that some bands in the sensitive cultivars but did not indicate tolerant one. On the other hand, the most tolerant cultivar Giza 2 showed a new band which was not found in any of the sensitive cultivars. These protein profile differences can be used as an indicator for susceptibility in maize cultivars against insect infestation. With regard to the date of cultivation, the four cultivars showed slight differences between the 1st and the 2nd plantation dates.

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التقدير المعمل للحساسية الموروثة لبعض أصناف الذرة ضد الإصابة بأفات ما بعد الحصاد

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اختبرت أربعة أصناف من محصول الذرة وهي: الذرة السكرية - الذرة هجين ثلاثى ٣٢١ - الذرة جيزة ٢ - الذرة هجين فردى ١٠ والتي تم زراعتها في ميعادين الأول في ٢٤ أبريل والثانى في ٢٨ مايو خلال موسم عام ٢٠٠١. واختبرت حساسيتها للإصابة بأفات ما بعد الحصاد معملياً. وقد لوحظ أن الحشرات الناتجة من الحبوب تابعة لفصيلتين من رتبتين مختلفتين هما فراشة الحبوب من رتبة حرشغية الأجنحة وسوسة الذرة من رتبة غمدية الأجنحة. وأظهرت النتائج أن الصنف جيزة ٢ هو أقل الأصناف إصابة بالحشرات من كلتا الفصيلتين كما وجدت فروق معنوية بين الأصناف الأربعة من حيث نسبة الإصابة بالحشرات و الفقد في وزن الحبوب ونسبة إنباتها. كما أمكن ترتيب الأصناف الأربعة تصاعدياً تبعاً لمعامل الحساسية للإصابة بالحشرات كالآتى: جيزة ٢ - هجين فردى ١٠ - الذرة السكرية - وكان الصنف الذرة هجين ثلاثى ٣٢١ هو أكثر الأصناف حساسية للإصابة. وبالتحليل الكهربى لبروتينات الأصناف تحت الاختبار أمكن الحصول على بعض الفروق في ترتيب الأحماض الأمينية تبعاً لوزنها الجزيئى مما يمكن معه التمييز بين الأصناف ومدى قابليتها للإصابة.