Minufiya J. Agric. Res. Vol. 34 No. 5:1913-1926 (2009) "http:// agri.menofia.edu.eg/mejla.html"

FIELD AND LABORATORY STUDIES TO EVALUATE THREE SQUASH CULTIVARS FOR THEIR RELATIVE SUSCEPTIBILITY TO SPIDER MITE *TETRANYCHUS URTICAE* KOCH AND THREE SUKING INSECT SPECIES

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(Received: May., 8, 2009)

ABSTRACT; *Greenhouse experiments were conducted to estimate the population fluctuation of the two spotted spider mite, Tetranychus urticae Koch, Thrips tabaci Lind., Aphids gossypii (Glov.) and Bemisia tabaci (Genn.) on three squash cultivars (American, Hitec and Eskandarani) at Qalubiea Governorate during a single growing season. Also, analysis of some specific chemical constituents of the leaves and clarified the resistance of the pest infestations.*

The obtained results indicated that the infestation by spider mite species, T. urticae was significantly different among the three different squash cultivars ($F_{2,128}$ = 10.49; P < 0.001). Eskandarani cultivar was the most susceptible harbored the highest average number of spider mite stages (609.90 individuals /leaf; LSD; P < 0.05). While Hetic cultivar was intermediate (483.62 individuals /leaf; LSD; P < 0.05). The lowest average number of spider mite infestations was recorded in case of American cultivar which was the lowest preferability to spider mite infestations (308.76 individuals /leaf; LSD; P < 0.02).

The results showed that significant difference was recorded on the leaves of different squash cultivars among the three phytophagous insect species ($F_{2,128}$ = 43.09 P < 0.05). Aphids gossypii was the highest number on all three squash cultivars (being highest on Eskandarani 12.61and Hitec 11.86 followed by American 11.21 individuals/leaf), followed by Thrips tabaci on Eskandarani 10.72, Hitec 9.45 and American 8.04 individuals/leaf; while the population of Bemisia tabaci was the lowest number on all squash cultivars (Eskandarani 1.24, Hitec 0.72 and American 0.16 individuals/leaf). However, there were no significant differences among the three different squash cultivars in leaf infested by insect species ($F_{2,128}$ = 1.17; p = 0.312).

The obtained data indicated that there were correlations between the phytochemical contents and the rate of mite infestation. There is a negative relationship between the infestation rate and the total phenol as well as the free amino acids contents in the squash cultivars, while there is a positive relationship between the infestation levels and the total sugar contents in

each cultivar. The American cultivar showed a lowest sugar content (12.02%) followed by the Hitec (12.95%) whereas, the Eskandarani had the highest content (16.55%), while the reverse was with the infestation rate with total phenol (0.35%, 0.28% and 0.23% for American, Hitec and Eskandarani, respectively). Also, the total free amino acids content resulted in a positive relation with the infestation rate for American (37.34%), Hitec (27.75%) and Eskandarani (22.43%). On the other hand, there were not correlations between the phytochemical contents and the rate of insect infestations.

Keywords: Phytophagous species, The two-spotted spider mite, Tetranychus urticae, Thrips tabaci, Aphis gossypii and Bemisia tabaci, Squash cultivars V., American, Hitec and Eskandarani, phytochemical contents

INTRODUCTION

Squash, *Cucrbita pepo* L. (Family: Cucurbitaceae) is one of the most important vegetables cultivated in both open fields and under greenhouses for local consumption and for exportation to the foreign markets. Squash cultivation is attacked by several phytophagous mite and insect species, which cause great economical losses to many crop species allover the world (Abdallah *et al.*, 2001; Ragkou *et al.*, 2004). The serious pest attacks plants, the two-spotted spider mite *Tetranychns urticae* Koch (Acari: Tetranychidae) feeds by sucking the plant sap (one mite can feed on 18 to 22 plant cells in 1 minute). This species covers the leaves with a mass of silken webbing (Abdallah, 2002). It has been recorded feeding on approximately 1200 described plant species in 70 genera (Ragkou *et al.*, 2004).

Estimation of the liability of squash to infestation with several pests especially under the greenhouses in order to select the most resistant one to avoid use any of chemical to control these pests. There were several studies on the host plant resistance to the infestation by *T. urticae* such as; Iskandar (2002) on pepper, El-Saiedy (2003) on two different strawberry, Abou-Zaid (2007) on five different cucumber cultivars.

They attributed this phenomenon to the chemical contents of plant tissue (Kielkiewicz *et al.* 1983) or to physical factors, leaf trichomes, morphological structure (Kielkiewicz *et al.* 1983). Induced resistance can result from an environmental change that may lead to temporary benefit of the host plant; the application of fertilizers, mineral nutrients can change the chemical constituents of plant tissue and consequently their nutritional value for pests (Ibrahim, 1988).

Painter (1951) defined tolerance as a resistance in which the plant shows an ability to grow and reproduce or to require pest injury to a marked degree in spite of supporting as insect population early equal to that damaging in susceptible host. Dahms (1972) identified 16 possible criteria to evaluate pest resistance in plants among which the number of pest motile stages attracted to cultivars when given a free choice.

The present investigations were carried out to evaluate the population fluctuation of phytophagous mite and insect species on three squash cultivars (American, Hitec and Eskandarani), and to analyze some specific chemical constituents of the following content: Total sugar, total phenol, and total free amino acids of their leaves.

MATERIALS AND METHODS

Three different squash cultivars (American, Hitec and Eskandarani) were cultivated under a greenhouse condition spider mite and insect infestations, during the season 2008, at Kafr El-Sohby village, Shebien El-Qanater Distrect, Qalubiea Governorate.

Five rows of each squash cultivars were planted alternatively and replicated three times for each one.

In order to study population fluctuations of the phytophagous species, leaf samples were collected weekly, starting on 8th of January until end of April. Fifteen leaves of each cultivar were randomly collected from each ofthree cultivars and placed directly into plastic bags and transported to the laboratory. All mite stages (eggs, immature and adults) and other insect pests were counted using stereomicroscope and the average number of mite stages per fifteen leaves were tabulated.

All the optimal agricultural practices were carried out for each of the cultivated plants. Also, the experimental area was kept free from any pesticide treatments. On the other side, the plants were left to the natural infestation and no artificial infestation was conducted.

One-way analysis of variance (ANOVA) and mean comparison using Fisher's least significant difference (LSD) were conducted for the number of phytophagous mite and insects, using the Super ANOVA program (Gagnon *et al.*, 1989). Significance level was $P \leq 0.05$.

The analysis of some phytochemical components:

Squash leaves of the three cultivars were collected and transferred to laboratory for analysis for the determination of the relationship among some phytochemical elements in the different squash cultivars and the levels of the infestation by phytophagous mite and insect species.

Leaves were cut to small pieces; five grams of these pieces for each cultivar were kept in small glasses which contain 50 ml ethyl alcohol 80 % at 10°C in the refrigerator. Samples were taken out of the refrigerator for homogenizing using a mixer. The homogenized samples were filtered through G-3 silica filter paper. Ethyl alcohol was added to the filterate up to 100 ml volume. Chemical analysis of some specific chemical constituents of the filterate was done according to the following procedures:

1- Total phenols determined by the colorimetric method of Floin-Denis as described by Swain and Hillis (1959).
2- Total amino acids were estimated as total of free amino acids according to Rosen (1965).
3- Total soluble sugars and total non soluble sugars were determined according to Smith *et al.* (1956).

RESULTS AND DISCUSSION

Three different squash cultivars namely (American, Hitec and Eskandarani) where chosen to study their relative susceptibility to phytophagous mite and insect species infestations during 2008 growing season, under greenhouse condition.

During the present study only the spider mite *T. urticae* and three insect *Thrips tabaci, B. tabaci* and *Aphis gossypii* species were found on the three squash cultivars. This work extends a similar study reported last years in which some plants were found to be immune to phytophagous species. The breeding of resistant cultivars is an important tool in integrated control programs.

The population fluctuation of *T. urticae* is recorded over the whole season (Table 1) and graphically illustrated (Fig. 1-a) during January to the end of April. Eskandarani cultivar was the most susceptible, where it's leaves harbored the highest population of spider mite stages (81.65 adults, 260.75 immatures, 267.50 eggs /leaf), followed significantly by Hitec cultivar (56.71 adults, 202.01 immatures, 224.90 eggs/leaf), while the American cultivar was infested by the lowest number of *T. urticae* stages (38.48 adults, 127.49 immatures, 142.78 eggs /leaf) wher as, American cultivars was resistant to spider mite *T. urticae* stages.

Squash infestations by *T. urticae* were early started and the mite population remained very lower throughout January and February. They started to increase rapidly to a peak in the mid March and then the population decreased rapidly. After that, they started to increase again towards the second peak at the end of April (Fig. 1-a). This increase may be attributed to increase in temperature during this period. Current findings agreed with previous studies, which showed that *T. urticae* life tables showed faster development as soon as temperature became higher (Wermelinger, 1990).

Many studies have revealed that photoperiod length and temperature are closely related to phytophagous population increases (Van de Vrie *et. al.*, 1972, Fujimoto and Takafuji, 1986).

Our study of three squash cultivars showed the existence of only one phytophagous mite, *T. urticae*, with very high populations and three phytophagous insect species; *Aphids gossypi, Thrips tabaci* and *Bemisia tabaci* with minimal populations.

Table 1: Number of different spider mite stages/leaf on three squash cultivars

Dates	American			Hitec			Eskandarani		
	Adult	Immature	Eggs	Adult	Immature	Eggs	Adult	Immature	Eggs
8-Jan	0.0	0.0	0.0	3.0	11.0	17.0	2.5	8.5	11.5
15-Jan	1.0	2.0	8.0	3.5	5.3	13.3	6.2	15.5	22.0
21-Jan	2.0	7.0	14.0	3.0	9.8	14.3	4.8	15.0	18.4
30-Jan	4.8	7.9	10.9	8.2	15.3	17.8	5.1	11.0	13.7
5-Feb	5.0	16.2	18.2	8.7	18.7	20.2	8.4	19.6	20.7
11-Feb	8.6	16.3	18.6	7.5	22.1	25.2	7.7	26.8	28.9
18-Feb	11.9	21.4	16.5	7.1	13.8	14.4	9.3	15.4	21.0
25-Feb	13.0	27.9	37.3	10.0	27.9	30.2	15.9	52.4	58.1
3-Mar	11.6	24.9	40.4	9.1	22.3	24.6	10.5	33.4	46.1
10-Mar	11.0	23.4	30.8	10.7	32.6	45.3	16.5	56.0	78.9
17-Mar	76.0	235.0	240.0	83.1	426.4	486.8	178.5	403.0	493.3
25-Mar	76.5	211.7	305.7	94.4	339.8	424.5	131.9	510.9	500.0
1-Apr	89.5	267.0	289.2	109.0	398.5	417.9	155.6	537.1	510.9
8-Apr	91.6	307.8	289.8	117.0	421.3	498.6	199.1	589.7	498.4
15-Apr	101.1	294.0	389.9	174.0	482.2	493.6	245.5	651.5	780.9
22-Apr	62.9	310.2	323.1	133.3	562.3	610.3	194.7	697.8	698.2
30-Apr	87.7	394.7	394.9	182.5	624.8	669.3	195.8	789.1	746.5
Mean	38.48	127.49	142.78	56.71	202.01	224.90	81.65	260.75	267.50
SE	9.67	34.90	37.86	15.89	56.97	62.15	22.46	72.94	73.15
Max.	101.10	394.70	394.90	182.50	624.80	669.30	245.50	789.10	780.90
Min.	0.00	0.00	0.00	3.00	5.30	13.30	2.50	8.50	11.50
Total average	308.76 $_c$			483.62 $_b$			609.90 $_a$		

Total average of each squash cultivars followed by different subscript letters is significantly different from each other ($P < 0.05$) LSD test

The results showed that significant difference was recorded on the leaves of different squash cultivars among the three phytophagous insect species ($F_{2,128}$ = 43.09 P < 0.05). However, there were no significant differences among the three different squash cultivars in leaf infested by phytophagous insect species ($F_{2,128}$ = 1.17; p = 0.312; Table 2).

The results concern the infestation of phytophagous insect species, *Thrips tabaci* were observed as follow: American 8.04, Hitec 9.45 and Eskandarani 10.72 individuals/leaf. Similar results were observed with the infestation of *Aphids gossypii* for the American, Hitec and Eskandarani cultivars (11.21, 11.86 and 12.61 individuals/leaf, respectively). The infestation of *Bemisia tabaci* was very low and recorded for American, Hitec and Eskandarani (0.16, 0. 72 and 1.24 individuals/leaf, respectively Table 2).

The infestation of the three cultivars by *Aphids gossypi* was started after mid January and increased (Fig. 1-b) then by the end of January decreased gradually, next they started to increase before the mid of March and reached their second peak by the end of March. Their number declined rapidly over a week and after that the population gradually increased to the third peak in the end of April for each cultivar (being: 44 to 50 individuals/leaf).

The infestation by *Thrips tabaci* was started in the end of January for Eskandarani and Hitec cultivars only (1.00 and 2.30 individuals/ leaf) then; it fell for a period till the second half of February and then they started to present for all squash cultivars and they began to increase sharply to a peak on 3rd of March, and after that declined for a week and they increased sharply again and reached their second peak by the end of April (being: 40 to 48 individuals/leaf; Fig. 1-c).

Bemisia tabaci was particula poor on the three squash cultivars. Squash leaves infested by *Bemisia tabaci* for month and half, infestation started from the end of January to by the mid of March. *Bemisia tabaci* were present in six sampling times for Eskandarani and Hitec cultivars (mean populations ranging from 0.72 to 1.24 individuals/ leaf), while this species was present only three times and disappeared for American cultivar (mean population 0.16 individual/ leaf; Table 2 and Fig. 1-d).

Generally, the rate of infestation by phytophagous mite and insect species on Eskandarani cultivar leaves were the highest about 1.5 to 2 times on these of Hitec and American cultivars respectively (Fig. 1). Eskandarani cultivar was highly susceptible cultivar, followed by Hitic cultivar. While, American cultivar resistance one.

Relationship between some phytochemical compounds and level infestation of spider mites and three species of insect,

One of the most important factors which may explain the susceptibility or the tolerance of squash cultivars to the infestation of phytophagous species is the phytochemical contents of their leaves.

Table 2: Number of insect pests /leaf on three squash cultivars

Date	*Aphids gossypii*			*Thrips tabaci*			*Bemisia tabaci*		
	American	Hitec	Eskandarani	American	Hitec	Eskandarani	American	Hitec	Eskandarani
8-Jan-08	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
15-Jan-08	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
21-Jan-08	1.10	8.10	12.20	0.00	0.00	0.00	0.00	0.00	0.00
30-Jan-08	1.20	7.10	11.00	0.00	2.30	1.00	0.00	1.00	2.10
5-Feb-08	0.50	4.10	4.40	0.00	0.00	0.00	0.00	1.50	1.40
11-Feb-08	1.80	2.00	2.45	0.00	0.00	0.00	0.00	1.40	1.60
18-Feb-08	0.00	2.00	4.40	0.00	1.09	2.64	0.98	4.32	7.14
25-Feb-08	1.80	2.90	5.60	0.00	1.24	3.00	0.60	2.86	6.90
3-Mar-08	2.14	3.53	3.57	0.65	1.38	3.46	1.13	1.20	1.93
10-Mar-08	0.44	1.26	1.85	0.98	1.53	2.56	0.00	0.00	0.00
17-Mar-08	1.00	2.00	2.14	1.50	2.40	3.14	0.00	0.00	0.00
25-Mar-08	11.20	8.90	14.53	7.98	11.41	10.31	0.00	0.00	0.00
1-Apr-08	8.11	9.30	8.40	6.11	10.73	11.38	0.00	0.00	0.00
8-Apr-08	17.70	24.50	14.80	16.10	18.22	25.90	0.00	0.00	0.00
15-Apr-08	43.70	37.30	30.06	21.70	27.80	24.20	0.00	0.00	0.00
22-Apr-08	49.67	38.55	54.30	40.90	44.60	48.40	0.00	0.00	0.00
30-Apr-08	50.18	50.16	44.70	40.80	37.90	46.18	0.00	0.00	0.00
Mean	11.21	11.86	12.61	8.04	9.45	10.72	0.16	0.72	1.24
SE	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Max.	50.18	50.16	54.30	40.90	44.60	48.40	1.13	4.32	7.14
Min.	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Total average	35.68$_a$			28.21$_b$			2.12$_c$		

Total average of each insect species followed by different subscript letters is significantly different from each other (P < 0.05) LSD test

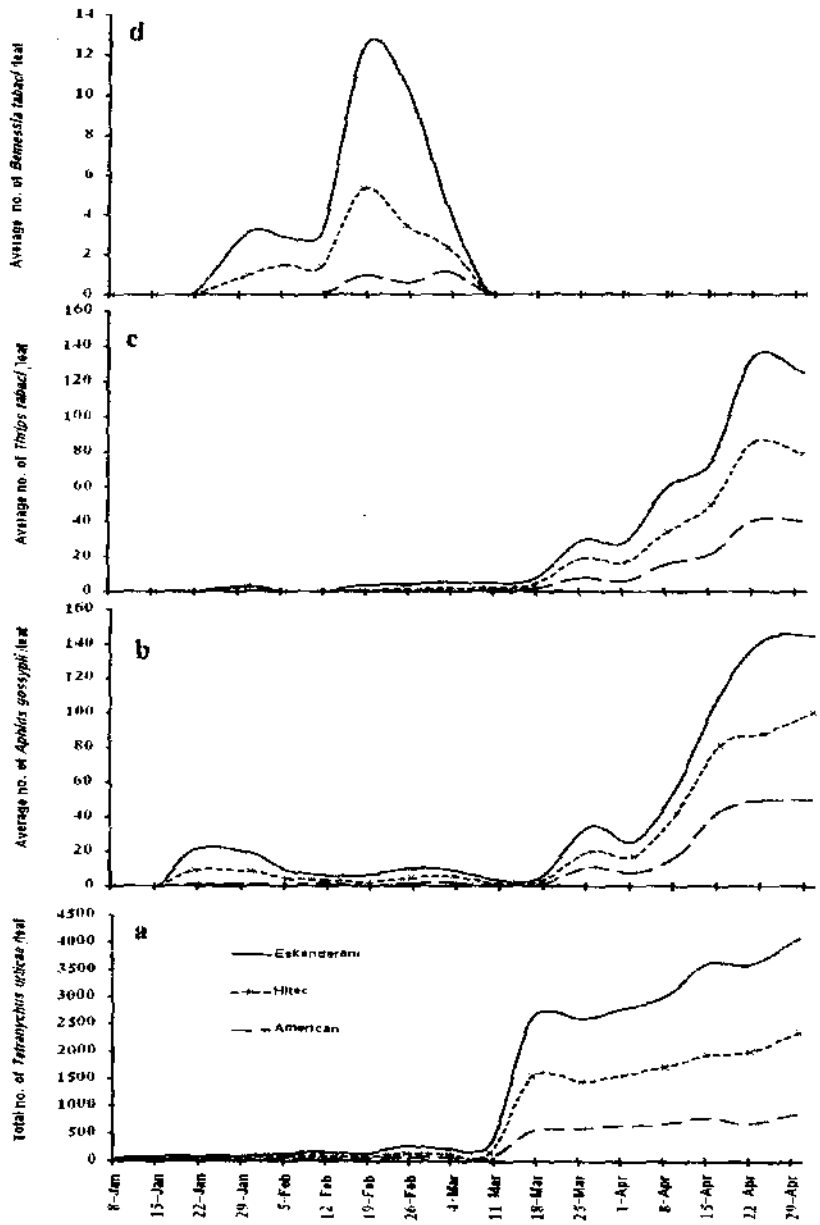

Figure 1: Total number of phytophagous mite and insects on leavies of the three squash cultivars

The levels of infestation were parallel with the plant content concentration. The highest mean number phytophagous mite species occurred on the leaves of Eskandarani cultivar (LSD; $P < 0.05$) was associated with highest levels of sugar content (16.55%; Table 3) and this indicates a positive significant relationship with the population densities. While the reverse was true with total phenol (0.23%) and total free amino acids (22.43%) indicated negatively significant relationships with the population densities as the results throughout the growing season.

The Hitec cultivar had moderate infestations (12.92%; Table 3), it showed moderate sugar content (12.95%), while the reverse was true with total phenol (0.28%) and total free amino acids (27.75%) compared with others. On the contrary, the American cultivar exhibited the lowest mean infestation number of phytophagous mite species (LSD; $P < 0.05$) and this was associated with lowest levels of sugar content (12.02%), while the reverse was true with total phenol (0.35%) and total free amino acids (37.34%) indicated negatively significant relationships with the population densities as the results throughout the growing season.

The obtained data indicated that there were correlations between the phytochemical contents and the rate of mite infestation. There is a positive relationship between mite infestation levels and total sugar contents in squash cultivars, while there is a negative relationship between the infestation rate and the total phenol as well as the free amino acids contents in each cultivar.

The obtained results are in agreement with that recorded by many authors; Kielkiewiez *et al.* (1983) stated that the phenol content distribution in the infested tissues of the resistance leaves is considered as an important factor in the defense reactions of plants against mite attacks. El-Saiedy (2003) and Mahgoub (2004) stated that, the mite resistance could be related to the high phenolic content in the infested leaves. Nel (1989) and El-Saiedy (2003) concluded that the high infestation of the mite may be related to the high sugar content exhibited.

It was evident from the above results that Eskandarani cultivar was the most suitable hosts for *T. urticae*. Contrary, American a poor host for *T. urticae*, we recommend growing the American cultivar which previously classified as the highest resistance one when compared with the two tested cultivars.

Table 3 : Chemical Composition of leaves for three squash cultivars during the season 2008.

Cultivars	Total Phenols (%)			Total free Amino acids g/100 g (%)			Total sugar (%)		
	Mean ± SD	Max.	Min.	Mean ± SD	Max.	Min.	Mean ± SD	Max.	Min.
American	0.35 ± 0.01	0.36	0.34	37.34 ± 0.78	38.22	36.74	12.02 ± 0.25	12.23	11.74
Hitec	0.28 ± 0.01	0.29	0.27	27.75 ± 0.73	28.56	27.15	12.95 ± 0.35	13.24	12.56
Eskandarani	0.23 ± 0.01	0.24	0.22	22.43 ± 0.66	23.18	21.96	16.55 ± 0.47	17.00	16.06

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دراسات حقلية ومعملية لتقييم ثلاثة أصناف كوسة للإصابة بالعنكبوت الأحمر *Tetranychus urticae* Koch وثلاثة انواع من الحشرات الثاقبة الماصة

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الملخص العربي

فى القليوبية اجريت تجربة بالصوبة لتقديرتذبذب تعداد الاصابة بالآفات الموجودة خلال موسم كامل على ثلاث انواع من الكوسة واجراء بعض التحاليل لمحتويات الاوراق وعلاقة بمقاومتها للآفات المختلفة.

واوضحت الدراسة تواجد نوع واحد من الأكاروسات المتغذية على النباتات *Tetranychus urticae* وبتعداد عالى جدا وثلاثة انواع من الحشرات المتغذيه نباتيا والمسماه*Aphis gossypii, Thrips tabaci* and *Bemisia tabaci*.

اظهرت النتائج للنوع الأكاروسى ان هناك اختلاف معنوى بين ثلاثة انواع الكوسة. حيث كان تعداد الأكاروس العنكبوتى عاليا على نوع الكوسة الاسكندرانى وهو اكثر الانواع حساسية حيث تحمل الورقة (حوالى 609.90 فرد)، و متوسطاً على نوع الكوسة الهيتك (حوالى 483.62 فرد / الورقة). وسجل اقل تعداد له على نوع الكوسة الامريكى(حوالى 308.76 فرد / ورقة) والذى يعتبر قل تفضيلا للاصابة بالأكاروس العنكبوتى.

اوضحت النتايج اختلافاً معنوياً بين الثلاثة انواع الحشرية على اوراق الكوسة المختلفة. حيث كان النوع الحشرى *Aphis gossypii* الاعلى تعداد على انواع الكوسة الثلاثة والتالى له النوع الحشرى *Thrips tabaci*. بينما تعداد النوع *Bemisia tabaci* كان

الاقل على الثلاثة انواع. وعلى الجانب الاخر لم يوجد اختلاف معنوى بين انواع الكوسة الثلاثة واصابتها بالانواع الحشرية.

قد وجد ارتباط بين المحتوى الكيماوى للورقه ومعدل اصابتها بالأكاروس. حيث توجد علاقة سالبة بين معدل الاصابة والفينول بالاضافة الى مركبات الاحماض الامينية فى اوراق الكوسة. وتوجد هناك علاقة موجبة بين مستويات الاصابة ومركبات السكر فى كل نوع من الكوسة . وسجل نوع الكوسة الامريكى الاقل فى عنصر السكر ١٢,٠٢% متبوعة بالنوع الهيتك، بينما يحتوى النوع الاسكندرانى الاكثر على ١٦,٥٥%. وعلى هذا الأساس فإن الصنف الأسكندرانى كان أكثر الأصناف قابلية للإصابة يلية الصنف الهيتك واقلها الأمريكى. وعلى النقيض، كان معدل الاصابة والفينول ٣٥. ، ٢٨. و ٢٣. % لكل من الامريكى، هيدك و الاسكندرانى بالترتيب. وايضا نتجت الاحماض الامينية علاقة موجبة مع معدل الاصابة لكل من الامريكى ٣٧,٣٤، الهيتك ٢٧,٧٥ والاسكندرانى ٢٢,٤٣. وعلى الجانب الاخر لا يوجد ارتباط بين محتويات الورقة الكيماوية ومعدل الاصابة بالانواع الحشرية المختلفه.