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Susceptibility of three squash cultivars to the two spotted spider mite, *Tetranychus urticae* (Acari: Tetranychidae) infestation in relation to phyto-chemical components of the leaves

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Abstract:

The field experiment was conducted to evaluate the relative susceptibility of three squash cultivars, *Cucurbita pepo* L. (Mabroka, Brencesa and Eskandarani) against the two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) during two successive years 2016 and 2017 in Balaktar and Om Saber villages, Behera Governorate. Also, it was carried out to study the effect of the morphological leaf characteristics and the chemical components (phenol, protein, chlorophyll and carbohydrate) of the three previously mentioned squash cultivars and clarified the resistance of the pest infestations. Obtained results showed that there was no significant difference in eggs or motile stages or the total numbers of spider mite infestation on the three cultivars *(*LSD; $P > 0.05$). However, Mabroka cultivar was the most resistant cultivar to the spider mite pest as its leaves harbored the lowest mean numbers *(*LSD; $P < 0.05$) and produced higher average of yield than that of Eskandarani cultivar in both villages, followed by Brencessa cultivar which occupied a moderate mean numbers of the spider mite in spite of produced higher total of yield production than that of Eskandarani cultivar. Which suffered the highest infestation of the spider mite population during both seasons, also its production was significantly the lowest average yield production ($P < 0.05$) in both locations. The morphological leaf characters, the number of in the lower surfaces were different in their shapes, length and number of setae for Mabroka, Brencesa and Eskandarani, which in 1 mm were 29, 39 and 61 setae, respectively, the results of chemical analysis of leaf component after pest infestation indicated positive relationships in case of total phenol (r = 1) for the three cultivars. While the amount of protein, chlorophyll and carbohydrates were significantly decreased and their (r) values were (-1) in the three cultivars. It could be classified to three degrees, the severe infestation as represented by (Eskandarani), the moderate infestation (Brencessa) and the last of low infestation (Mabroka). It is concluded that growing the Mabroka cultivar better than the other two tested cultivars.

Introduction

Squash, *Cucrbita pepo* L. (Family: Cucurbitaceae) has a high nutritional value essential for the body metabolism and vitality as they contain different vitamins, sugar, starch, fats, proteins and minerals such calcium magnesium, potassium, sodium, phosphorus and iron. The squash plants are attacked by the two spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) which infest leaves, stems, branches and causing various degrees of damage, they usually feed on the leaves injuring the epidermis resulting in blotching, stippling or bronzing and consequently reduce quantitatively and qualitatively the yield. (Faris *et al.,* 2004 and Abdallah *et al*., 2009). The infestation with the spider mites may be affected by plant leaf morphological structure and its chemical contents (El-Saiedy *et al.,* 2011). Leaf structure has been showed to be related to mite damage or to symptoms commonly associated with damage (Abou-Zaid, 2013). The concentration of organic compounds in the tissue of plant leaf i.e. proteins, carbohydrates, phenolic compounds can be influenced by mite feeding as the reduction of total protein was occurred, the injured leaves protein can be degenerated or its synthesis is prohibited. In addition, changes in the concentration of soluble sugars and amino acids not only create better physical conditions for feeding but also provide a higher nutritive value of the food for spider mites. (Tomczyk and Kropczynska, 1985) Many studies were done to clarify the relation between mite infestation and leaf structure. Some varieties of Pelargonium with thicker cuticle and epidermis were more resistant to spider mite. Besides that, the upper leaf surface was less preferred by mites, because its cuticle and epidermis were, thicker than that of the lower surface (Kou *et al.,* 1972 and Luczynski *et al.,* 1990). Kielkiewicz (1994) and Magali (1997) stated that, leaf trichomes contributed to the reduction of *Tetranychus cinnabrinus* (Boisdaval) (Acari: Tetranychidae) density at beginning of mite feeding and on tomato and bean varieties. Adults of *T. urticae* could feed through the spongy and part of the palisade parenchyma of the leaf, while immature *T. urticae* could feed only through the spongy parenchyma (Park and Lee, 2002). Also, the length, density and thickness of leaf trichomes may be considered as other factors affecting the host plant resistance to infestation by *T. urticae* (El-Saiedy, 2003). A negative relationship between the thickness of the upper epidermis and spongy tissue of leaf of the 7 cucumber varieties and the abundance of *T. urticae* movable stages. While, it was positive in case of palisade tissue (Hanafy, 2004). Moreover, *T. urticae* preferred the lower leaf surface of the plants due to the very thin cuticular layer (Abo-Bakr and Ali, 2005). Also, the more thickness of the cuticle of the epidermis especially that of the lower surface could be considered as a physical resistance factor (Azouz, 2005). Resistant varieties possess thicker on either upper or lower leaf surface than susceptible ones. The more thickness of the cuticle epidermis especially that the lower surface could be considered as a physical tolerance factor against mechanism of spider mite (El-Sanady *et al.,* 2008). The present study was carried out as a trial to throw light on the susceptibility of the three squash cultivars to spider mite's infestation in realtion to phyto-chemical components of the leaves

Materials and Methods

1. Field studies:

1.1. Population study of the *Tetranychus urticae*:

The experiment was conducted in Balakter and Om-Saber villages, El Beheira Governorate to evaluate the population and the susceptibility of three squash cultivars (Mabroka, Brencessa and Eskandarani) to the spider mite infestation, *T. urticae*. An area of 350 m^2 for each cultivar was divided into four equal plots each of 87.5 m^2 (one plot for cultivar sensitivity and three plots for measurement

of different methods of control). Seeds of the three cultivars were sown on 11th& 14th of March in 2016 and 2017 seasons, respectively. Each plot consists of 10 ridges, 70 cm apart, 12 m long and thinned to 2 plants in the raw to give a population of 2400 plants per 1/4 feddan. The experimental design was a randomized complete block with three replications. All plots received the recommended agricultural practices.

Samples of randomly chosen leaves from the squash plants were picked up weekly from each cultivar after three weeks of cultivation until the end of the season (30 leaves / cultivars, continued for 17 weeks) and each sample were kept in a tight closed plastic bag. After that they were transferred to the laboratory for examination using stereomicroscope. Eggs and movable stages (larva, nymphs and adults) of the two-spotted spider mite attacked the leaf surface were estimated by counting their numbers on each leaf for the tested squash cultivars during growing stages.

2.Laboratory studies

2.1. Effect of chemical components and leaf anatomy on the density of *Tetranychus urticae* on the three cultivars of squash:

2.1.1. Effect of *Tetranychus urticae* infestation on the phyto-chemical components: of leaf:

Chemical analysis of leaf samples was carried out at the beginning (seedling stage) and at the peak of spider infestation (fruiting stage). Samples of squash leaves of the three cultivars were transferred to the Faculty of Agriculture Research Park, Cairo University for chemical analysis. Some specific chemical constituents of squash leaves cultivars were determined as follow: Total phenol content was determined by Folin-Ciocateu method as modified by Singelton and Rossi (1965). Total carbohydrates were extracted from the plant leaves and prepared for assay according to Crompton and Birt (1967). Total protein was calorically assayed by ninhydrin reagent according to the method described by Lee and Takabashi (1966). Total chlorophyll was determined calorimetrically according to Holden (1965).

2.1.2. The leaf anatomy (morphological leaf characteristics):

Leaf samples of three squash cultivars were collected and imaged the lower surface of leaves using the Analytical Scanning Microscopic Technique (SEM) at the National Research Center according to (Karnousky, 1965 and Fischer *et al*. 2012). Trichomes were counted by using Computer Eye.

3. Data analysis:

Analysis of variance was conducted by one way to determine the significance between means of the tested cultivars by using the portable statistical analysis SAS 9.3.1. program (SAS Institute, 2003). Whereas the means were compared through LSD tests, least significant differences at 0.05 level. Mean numbers of the two-spotted spider mite, *T. urticae* on each squash cultivar (Mabroka, Brencessa and Eskandarani) in each village were compared using Student's t-test. The simple Correlation coefficient was also used by using Pearson Simple Correlation Coefficient Calculator*

Results and Discussion

1.Population of the two-spotted spider mite, *Tetranychus urticae* on the three tested cultivars:

The population abundance of the two-spotted spider mite, *T. urticae* egg and motile stages (immature stages and adults) on the three squash cultivars (Mabroka, Brencessa and Eskandarani) were studied during 2016 and 2017 seasons in Balaktar and Om Saber villages, Behera Governorate. Figures (1 and 2) showed the relation between time (week) and the total average numbers of the spider mite (individual) for the three cultivars. In the first season, the spider mite started on the leaves of the three squash cultivars (Mabroka, Brencesa and Eskandarani) on the

3rd of April in smaller numbers and the mite population remained very lower for the first three weeks from the beginning of the experimental survey. Then the total number of spider mite populations increased gradually through the next four weeks on 22nd of May and reached approximately 38.68, 47.51 and 59.14 individual/leaf; respectively in Balakter village and 90.66, 159.26 and 177.44 individual/leaf; respectively in Om Saber village. After that the spider mite populations continued to increase rapidly in numbers for the rest of the season reaching at the end of the season 216.22, 568.28 and 751.22 individual/leaf in Balakter village and 330.46, 815.35 and 1176.28 individual/leaf; respectively in Om Saber village (Figure, 1). In the second season**,** likewise the total number of spider mite on the three cultivars (Mabroka, Brencesa and Eskandarani) revealed the same trend and started on 31st of March in a small number as well and the curves were approximately consent for the first three weeks from the beginning of the experiment. Then the populations numbers increased and reached approximately 43.71, 54.42 and 65.24 individual/leaf; respectively in Balakter village and 73.07, 129.41 and 158.52 individual/leaf in Om Saber village, through the next four weeks on 14 of May and then continued to increase rapidly in numbers and this increase continued till the end of the season at which the population reached 245.41, 597.1 and 768.25 individual/leaf in Balakter village, while the total number of spider mite populations in Om Saber village reached to 355.45, 832.21 and 1213.24 individual/leaf; respectively (Figure, 2).

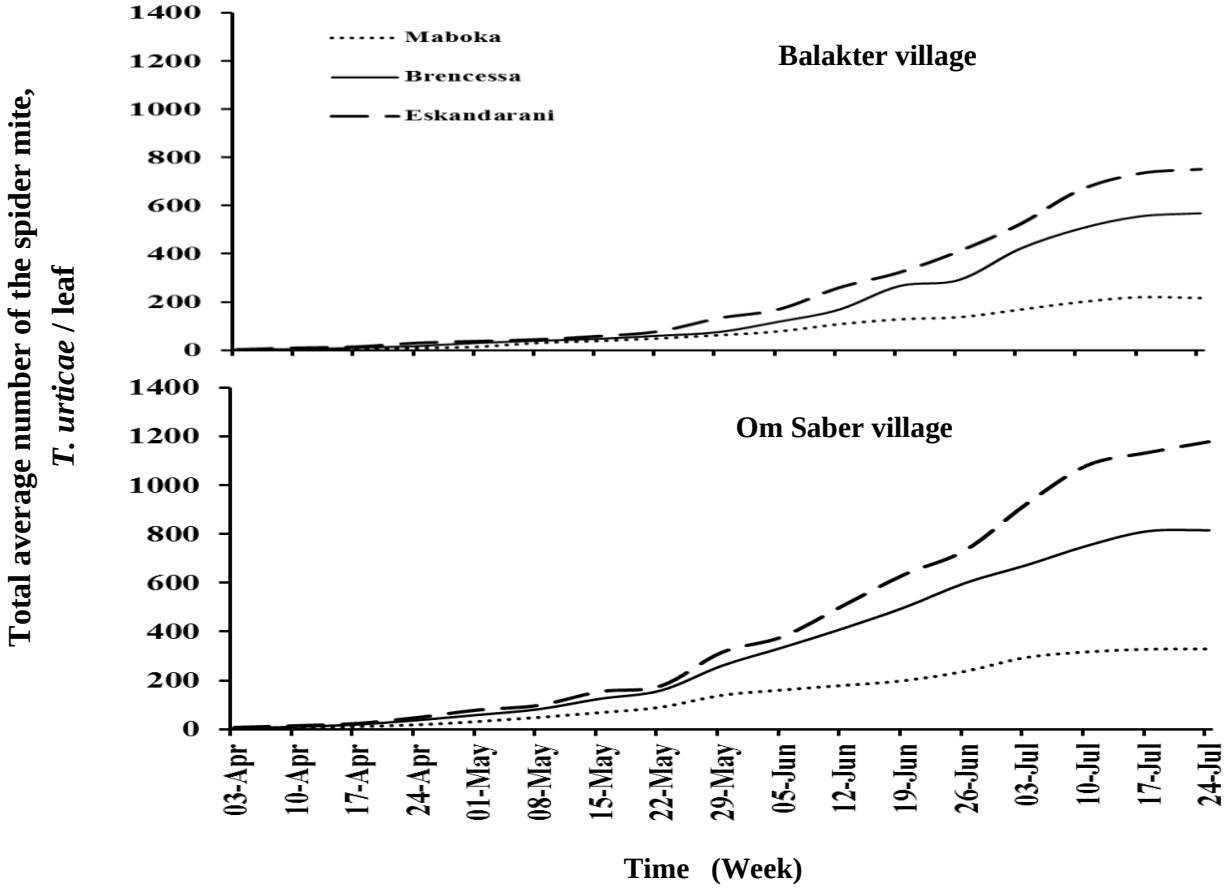


Figure (1): Weekly total average numbers of the spider mite, *Tetranychus urticae* /leaf on three squash cultivars; Mabroka, Brencessa and Eskandarani; under field conditions in Balakter and Om Saber villages, Behaira Governorate during season 2016.

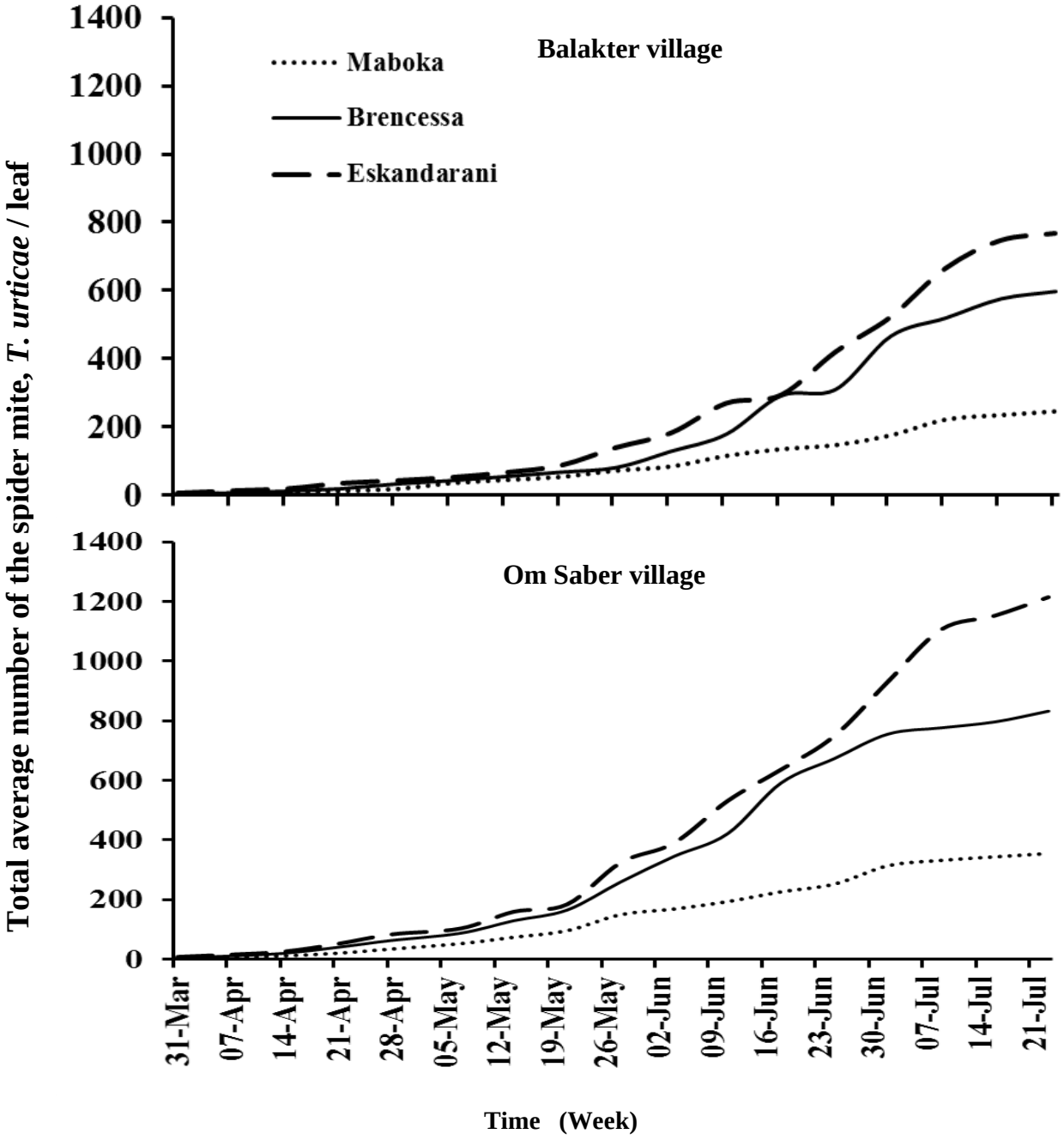


Figure (2) : Weekly total average numbers of the spider mite, *Tetranychus urticae* /leaf on three squash cultivars; Mabroka, Brencessa and Eskandarani; under field conditions in Balakter and Om Saber villages, Behaira Governorate during season 2017.

Table (1) described the average number for the egg and motile stages (immature stages and adults) as well as the total of spider mite, *T. urticae* ± standard error (SE), Maximum and on Minimum numbers on Mabroka, Brencessa and Eskandarani cultivars throughout 2016 and 2017 seasons in Balaktar and Om Saber villages.

Table (1): Total average numbers of the two-spotted spider mite, *Tetranychus urticae* /leaf of squash cultivars; Mabroka, Brencessa and Eskandarani under field conditions at Balaktar and Om Saber villages during both seasons.

Number of spider mite stages	Squash cultivars	2016					
		Balakter			Om Saber		
		Mean ± SE	Max.	Min.	Mean ± SE	Max.	Min.
Eggs	Mabroka	42.47 ± 9.07$_{aA}$	106.41	0.90	78.45 ± 16.95 $_{aB}$	190.14	0.40
	Brencessa	82.19 ± 20.60$_{abA}$	236.11	1.94	133.67 ± 26.45$_{abA}$	300.11	2.35
	Eskandarani	108.51 ± 27.41$_{bA}$	325.11	2.16	179.47 ± 39.17$_{bA}$	475.14	4.81
Motile stages	Mabroka	44.1 ± 9.97 $_{aA}$	113.55	0.14	66.82 ± 12.85 $_{aA}$	141.17	0.23
	Brencessa	105.04 ± 29.45$_{abA}$	332.17	0.98	199.18 ± 47.40$_{abA}$	515.24	1.88
	Eskandarani	141.85 ± 37.88$_{bA}$	427.11	1.75	260.54 ± 64.25$_{bA}$	701.14	3.11
Total	Mabroka	86.57 ± 19.00$_{aA}$	219.96	1.04	145.27 ± 29.69$_{aA}$	330.46	0.63
	Brencessa	187.22 ± 49.99$_{abA}$	568.28	2.92	332.85 ± 73.56$_{abA}$	815.35	4.23
	Eskandarani	250.36 ± 65.24$_{bA}$	751.22	3.91	440.01 ± 103.21$_{bA}$	1176.28	7.92
		2017					
Eggs	Mabroka	46.06 ± 9.91 $_{aA}$	122.15	1.11	81.8 ± 17.35 $_{aB}$	195.13	0.48
	Brencessa	86.00 ± 21.42$_{abA}$	248.90	1.82	136.97 ± 26.50$_{abA}$	300.10	2.60
	Eskandarani	111.42 ± 27.89$_{bA}$	330.14	2.20	184.07 ± 39.89$_{bA}$	481.13	4.32
Motile stages	Mabroka	47.83 ± 10.75$_{aA}$	123.26	0.18	72.59 ± 14.28$_{aA}$	160.32	0.14
	Brencessa	112.97 ± 31.09$_{abA}$	348.20	1.11	214.39 ± 51.22$_{abB}$	532.11	1.92
	Eskandarani	142.32 ± 37.83$_{bA}$	438.11	2.00	266.06 ± 65.68$_{bA}$	732.11	3.00
Total	Mabroka	93.88 ± 20.62$_{aA}$	245.41	1.29	154.39 ± 31.54$_{aA}$	355.45	0.62
	Brencessa	198.97 ± 52.45$_{abA}$	597.10	2.93	351.36 ± 77.32$_{abA}$	832.21	4.52
	Eskandarani	253.75 ± 65.6$_{8bA}$	768.25	4.20	450.13 ± 105.38$_{bA}$	1213.24	7.32

Means followed by different subscript small letters within columns for mite stages are significantly different from each other. Also, Means in row followed by different subscript capital letters within the row are significantly different from an another year (P < 0.05) LSD test

As shown in Table (1) no significant difference was found in the spider mite population among the three cultivar leaves (LSD; $P > 0.05$) during seasons 2016 and 2017 in both Balaktar and Om Saber villages. In the first season, there was significant difference in the number of spider mite eggs on Mabroka cultivar only between Balakter and Om Saber villages (LSD; $P < 0.05$). However, there were no significant differences between both villages in number of spider mite eggs or motile stages or total on the leaves of other cultivars (LSD; $P > 0.05$; Table, 1). There was no significant difference in eggs or motile stages or the total number of spider mites on the three cultivars (LSD; $P > 0.05$) (Table, 1). However, each stage of the spider mite on the three cultivars was ranked in ascending order on the leaves of Mabroka, Brencessa and Eskandarani cultivars during the two seasons in both villages. The total average number of the spider mite on Eskandarani cultivar was about folds that in Mabroka cultivar (250.36 and 86.57 individuals/ leaf, respectively in Balakter village; also, there were 440.01 and 145.27 individuals/ leaf, respectively in Om Saber village (Table, 1). While Brencesa cultivar had a moderate total average number of the spider mite 187.22 individuals/leaf among the tested cultivars in their infestation by *T. urticae*; in Balakter village and 332.85 individuals/leaf in Om Saber village (Table, 1). In the second season**,** similar results were

found in the infestation of the three cultivars under test with the spider mite, which were in the same direction as they were observed in both villages, except Brencessa cultivar which was a significant difference in the population number of motile stages on the leaves between both villages (LSD; $P < 0.05$) (Table, 1).

Moreover, the total average number of the spider mite on Eskandarani cultivar was about doubles that in Mabroka cultivar (253.75 and 93.88 individuals/leaf, respectively in Balakter village; also there were 450.13 and 154.39 individuals/ leaf, respectively in Om Saber village (Table, 1). While Brencesa cultivar had a moderate total average number of the spider mite 198.97 individuals/leaf among the tested cultivars in their infestation by *T. urticae*; in Balakter village and 351.36 individuals/leaf in Om Saber village (Table, 1). This cultivar was not significantly different from Mabroka or Eskandarani in both villages (LSD; $P > 0.05$).

The infestation rate by *T. urticae* in Om Saber village was higher than in Balakter and Om Saber village, with no significant difference between them (Figure, 3). Eskandarani was the most susceptible cultivar as its leaves harbored the highest total of spider mite population (250.36 and 440.01 individuals/leaf; in Balaktar and Om Saber villages, respectively during season 2016 and 253.75 and 450.13 individuals/leaf; in Balaktar and Om Saber villages, respectively during season 2017 (Table,1), followed not significantly by Brencessa cultivar (187.22 and 332.85 individuals/leaf; in Balaktar and Om Saber villages, respectively during season 2016 and 198.97 and 351.36 individuals/leaf; in Balaktar and Om Saber villages, respectively during season 2017, while the Mabroka cultivar was infested by the lowest total of spider mite population (86.57 and 145.27 individuals/leaf; in Balaktar and Om Saber villages, respectively during season 2016 and 93.88 and 154.39 individuals/leaf; in Balaktar and Om Saber villages, respectively during season 2017 (Table, 1 and Figure, 3), which is the most resistant one to the total of spider mite population. This result coincides with that obtained by Allam (2014) who studied the susceptibility of four squash varieties to spider mites and other different phytophagous pests' infestation and he concluded that the highest population of mite was recorded in variety of Eskandarani (baldy) and in contrary, Mabroka variety exhibited less population of the two-spotted spider mite.

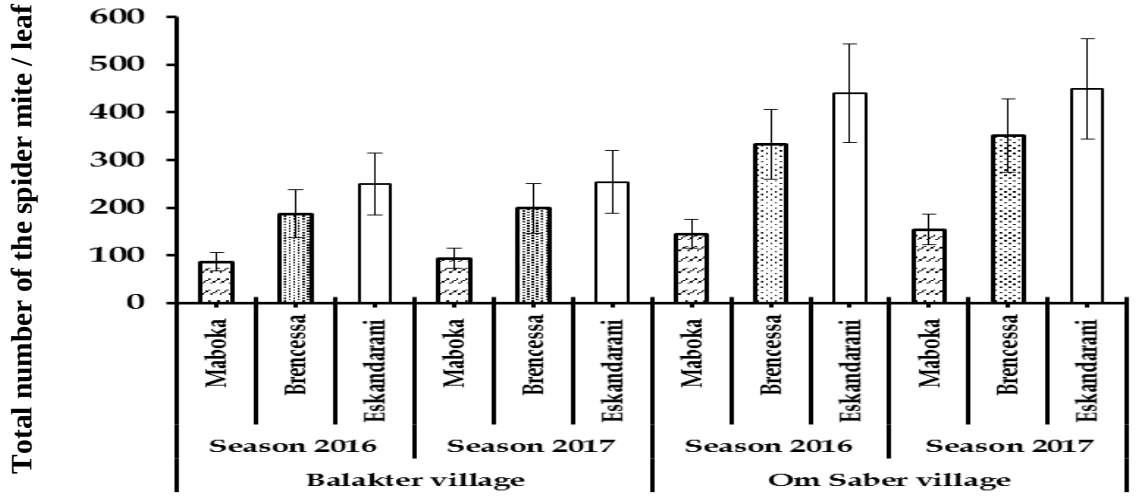


Figure (3): The total number of the two-spotted spider mite, *Tetranychus urticae* on the squash leaf of the three cultivars in Balakter and Om Saber villages in both seasons.

2. Measurements yielding of each squash cultivar:

Total of squash yields for each cultivar was collected and weighted to estimate the final yield per two karats. Table (2) described the average of the three squash yields in kg. /2 Karats, the mean of yield production, Maximum and on Minimum yield of Mabroka, Brencessa and Eskandarani cultivars in Balaktar and Om Saber villages. Data in that table showed the highest total of yield production were obtained of Brencessa cultivar recorded (835.76 and 707 Kg / 2 karats) in Balaktar and Om Saber villages, respectively, followed by Mabroka cultivar (664.6 and 585.4 Kg / 2 karats) then the lowest total of yield production were obtained for Eskandarani cultivar (446.86 and 362.5 Kg / 2 karats (Table, 2). There was significant difference among the three squash cultivars in total yield production/2 karats (LSD; $P < 0.05$) in both villages. Also, there were no significant difference between Brencessa and Mabroka cultivars in the mean of yields. However, there was a high significant difference between Mabroka and Eskandarani cultivar, which produced the lowest amount of yield production (LSD; $P < 0.05$) (Table, 2 and Figure, 4) in both locations.

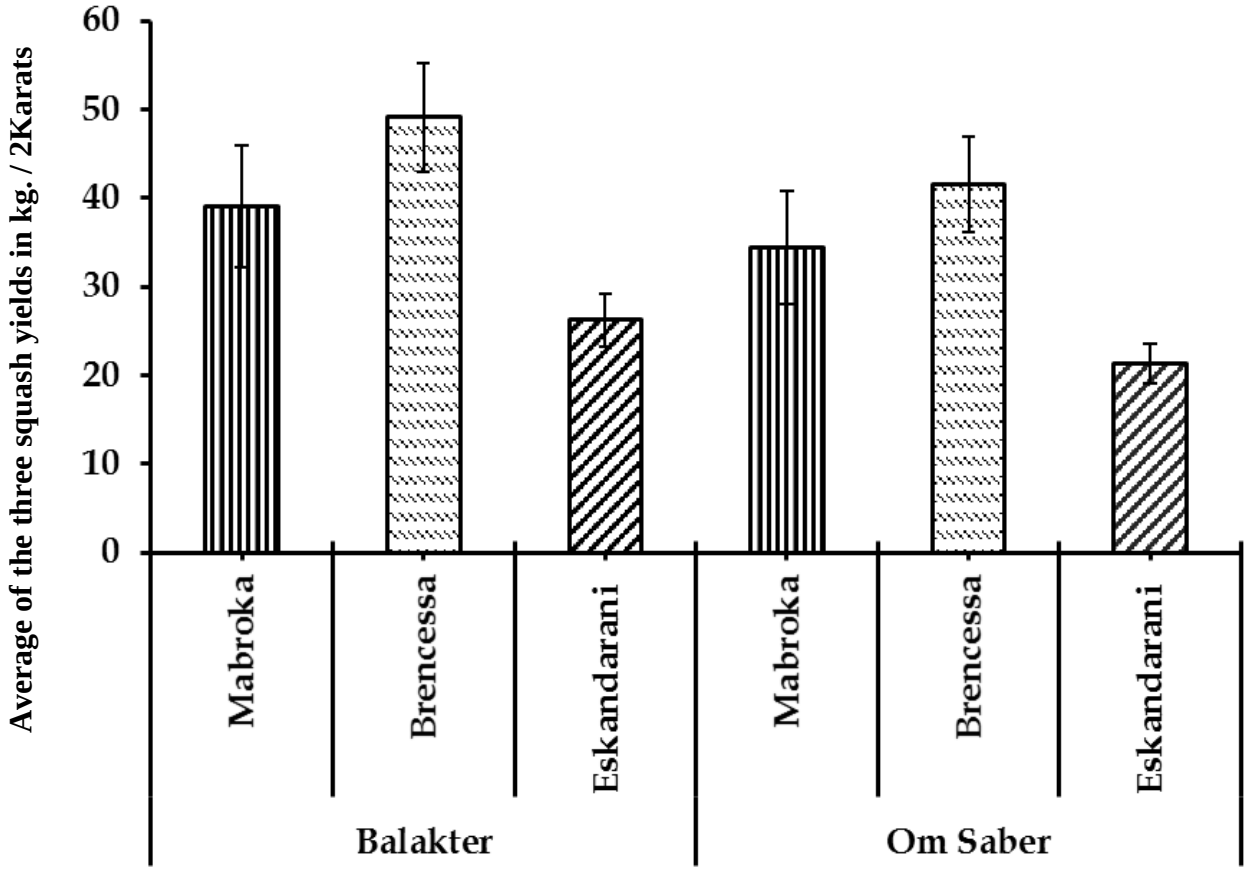


Figure (4) : Average of the three squash yields in kg. / 2Karats in both villages.

Table (2): Total of the three squash yields (Mabroka, Brencessa and Eskandarani) in kg. / 2 Karats in Balaktar and Om Saber villages.

Date	Balakter village			Om Saber village		
	Mabroka	Brencessa	Eskandarani	Mabroka	Brencessa	Eskandarani
25-May	8.8	42.3	14.5	5.7	38	10.4
29-May	13.2	20.6	20.8	9.5	17	16.7
02-Jun	20.3	24.94	9.4	17.6	20.3	7
06-Jun	70.9	16.54	13.8	65.4	12.7	10.8
10-Jun	43.4	40.8	20.5	39.3	35.4	17.4
14-Jun	96.7	70.9	35	83.7	55	30.2
18-Jun	50.6	90.3	21.5	47.4	73.8	17.5
22-Jun	20.4	76.94	40.2	19	67.3	33.6
26-Jun	32.3	81	61.4	28.3	79	41.5
30-Jun	30	51.54	31	27	46	27.3
04-Jul	14	71	27.96	10.8	53	23.4
08-Jul	18	41.3	30.9	13	37.3	25.2
12-Jul	10	44.8	21.5	7	39.7	18
16-Jul	26	87	29.6	20.2	73.5	24
20-Jul	50	21.8	30.8	43.5	18	26.5
24-Jul	90	30	22	82.7	24	19.3
28-Jul	70	24	16	65.3	17	13.7
Total	664.6	835.76	446.86	585.4	707	362.5
Mean	39.1_{ab}	49.2_{a}	26.3_{c}	34.4_{ab}	41.6_{a}	21.3_{c}
Max.	96.7	90.3	61.4	83.7	79	41.5
Min.	8.8	16.54	9.4	5.7	12.7	7.0

Means followed by different subscript letters within row are significantly different from each other (P < 0.05) LSD test.

3. The laboratory studies:

3.1.Effect of some chemical components of squash leaves and their relation to spider infestation:

The two-spotted spider mite, *T. urticae* infested the three tested cultivars of squash with different rates. Data in Table (3) showed the nutrient content of the tested squash cultivars in order to investigate the relationship between components and the mite pest infestation. The chemical analysis of leaf samples was carried out at the beginning and during the peak season of infestation.

Table (3): Some chemical components of squash leaves and their relation to spider infestation during season 2017.

Cultivars	Stage of plant	Mean No of spider mite	Total Phenol	Total protein	Total chlorophyll	Total carbohydrate
Mabroka	Seedling	4.56	12.05	23.96	122.54	14.68
	Fruiting	113.55	15.11	20.44	117.77	11.66
R			1	-0.99	-1	-0.99
Brencessa	Seedling	8.6	10.98	20.98	121.33	13.91
	Fruiting	321.11	12.82	18.95	114.65	10.23
R			1	-1	-1	-1
Eskandarani	Seedling	14.34	9.14	21.85	124.55	14.87
	Fruiting	427.11	11.58	18.23	113.22	10.85
R			1	-1	-1	-1

R: correlation coefficient

The three cultivars varied significantly in their infestations with the spider mite as the individuals were more abundant on plant leaves of Eskandarani followed by Brencessa while the lower abundant of the pest occurred in Mabroka.

Regarding Mabroka, it was noticed that at the beginning of infestation, when the population density of mites is minimum 4.56 individuals /leaf the value contents of protein, chlorophyll and carbohydrate were 23.96, 122.54 and 14.68 mg/100 gm, respectively; these values were decreased to 20.44, 117.77 and 11.66 mg /100 gm, respectively, when the population density of mites was in its peak of infestation recorded 113.55 individuals /leaf. While for total phenol throughout the beginning of infestation, recorded a low value 12.05 mg /100 gm, which increased to 15.11 mg /100 gm when the population density of mites was maximum. Concerning the relation between the population levels and the previously mentioned components, the calculated correlation coefficient values were significantly positive in case of 2total phenol as the corresponding r values is (1), while reversal relationships were detected with protein, chlorophyll and carbohydrate as the correlation coefficient values were -0.99, -1 and -0.99, respectively.

Concerning Brencessa; it was clear that its leaves were infested with 8.6 and 321.11 individuals /leaf of the spider mite at the beginning and at the peak of infestation, respectively, and by evaluating some of the phytochemical components in Brencessa leaves; the result revealed that it contained 20.98, 121.33 and 13.91 mg /100 gm of protein, chlorophyll and carbohydrate at the beginning of infestation which decreased at the end of infestation and became 18.95, 114.65 and 10.23 mg /100 gm of the previous components, respectively. Meanwhile, for the repellent component, the phenol, the data showed a low value 10.98 mg /100 gm at the beginning of infestation when compared to those of peak infestation, 12.82 mg /100 gm. Statistically, the correlation coefficient clearing the fact by increasing the population density of the spider mites on cultivar leaves; its contents of protein, chlorophyll and carbohydrate decreased significantly as their values were (-1) of the three compounds, while total phenols affected positively on population density of the spider mites i.e. the population increased by increasing the content of phenols as the correlation value was (1).

With respect to Eskandarani, which classified as the highest susceptible one of the three cultivars, it was observed that its leaves were infested with 14.34 and 427.11 individuals/leaf throughout the beginning and at the peak of infestation. Its leaves possessed a high value of nutrient contents recorded 21.85, 124.55 and 14.87 mg /100 gm of protein, chlorophyll and carbohydrate, respectively, during the beginning of infestation and those components were decreased by increasing the population of the pest and reach its peak which became 18.23, 113.22 and 10.85 mg /100 gm. While total phenols were 9.14 mg /100 gm when the population of the pest at the beginning of infestation which increased to 11.58 mg /100 gm at the end of infestation.

By concerning the correlation coefficient calculated to clear the relation between the densities of spider mites and those components. Results indicated positive relationships in case of total phenol (r = 1), while its defense secretion increased in the cell sap. On the other direction by increasing the population density of spider mites, the amount of protein, chlorophyll and carbohydrates were significantly decreased, their r values were (-1) in the three components.

These results revealed that the lowest variety infested by spider mite was Mabroka. The chemical analysis showed that this cultivar had a high content of phenol and protein at the beginning of infestation (12.05 and 23.96 mg /100 gm, respectively) compared to the highest variety of infestation (9.14 and 21.85 mg /100 gm) of (Eskandarani) at the end of the season.

Data showed that Mabroka cv. was the lowest squash cultivar to spider mite infestation. It had the least number of motile stages of the pest (113.55 individuals) at the peak infestation. The second rank of mite infestation was occupied by Brencessa which its corresponding value was 321.11 individuals of the pest. However, the third rank of infestation recorded the highest pest infestation (427.11 individuals) belonged to Eskandarani cv. i.e. an increase in the population density of spider mite, the amount of protein, chlorophyll and carbohydrates were significantly decrease, while an increase in the amount of phenol, the cultivar acquires resistance and the pest the spider mite decreases. This result was coincided with that of Tomczyk and Kropczynska (1985) they proved that proteins, carbohydrates, phenolic compounds can be influenced by mite feeding as the reduction of total protein was occurred, the injured leaves protein can be degenerated or its synthesis is prohibited. In addition, changes in the concentration of soluble sugars and amino acids not only create better physical conditions for feeding but also provide a higher nutritive value of the food for spider mites. Also, Tomczyk *et al.* (1987) studied the amino acids and soluble proteins in resistance of four cucumber to the tetranychid mite, *T. urticae*. They indicated that the increases in photosynthetic activity and growth in plants as a result of mite feeding sometimes led to increase in yield. Moreover, Ahmed (1994) proved that the susceptible cucumber cultivars to infestation with *T. urticae* contain more protein, total amino acids, than resistant varieties. In addition, Mahgoob (2004) studied the effect of chemical constituents and anatomical structure of some host plants leaves against the two-spotted spider mite *T. urticae*. He observed that the mite resistance could be related to the high phenolic content as in *Dodonaea viscosa* and *Citrus aurantium*. Taha and EI-Raies (1996) showed that, the increasing of salinity levels increased leaf sodium, and decreased nitrogen, in consequence, the mite infestation was decreased. Abdallah *et al.* (2009) studied the correlation between the phytochemical contents and the rate of mite infestation higher number of phytophagous mites which was associated with higher levels of sugar content and this indicates a positive significant relationship with the population densities and sugar content. While the reverse was true with total phenol (which indicates a negative significant relationship with the population densities. El-Saiedy *et al.* (2011) stated that infestation with the spider mites may be affected by its chemical contents; a positive relationship occurred between mite infestation levels and total soluble sugars in watermelon cultivars while negative relationship found with total phenol compounds.

3.2. Effect of leaf anatomy on the density of spider mite on the three cultivars of squash:

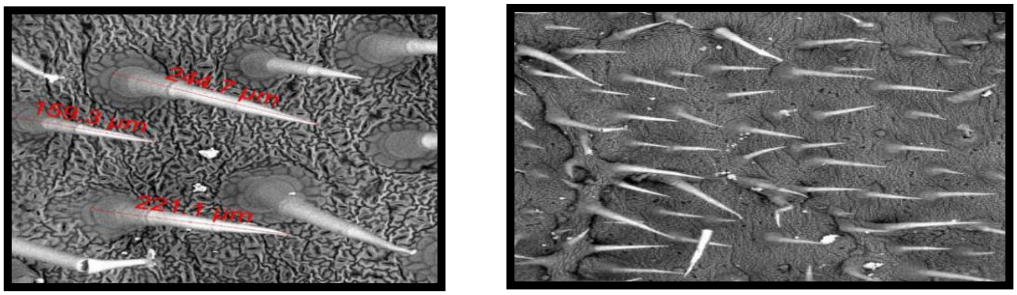
The susceptibility of squash plants to infestation with affected by plant leaf morphological structure. Therefore, the morphological leaves of the three squash cultivars, Mabroka, Brencessa and Eskandarani were studied (Figure, 5).

The number of trichomes (Table, 4) in the lower surfaces in mm were 29, 39 and 61 setae for Mabroka, Brencessa and Eskandarani, respectively. The length of the longest seta was 285.2 µm, for Brencessa followed by 267.4 µm, for Eskandarani and then 174.6 µm for Mabroka. The length of the smallest seta was in Eskandarani of 37.5 µm followed by Mabroka of 50 µm and then Brencessa of length 56.25 µm; setae of Mabroka spine in shape, short with 2 segments; setae of Brencessa, flagellate with two segments; setae of Eskandarani flagellate with three segments. The anatomical structure of some host plants leaves of three cucurbit crops, Sudanian watermelon, snake watermelon and squash on their susceptibility to the spider mite infestation (Ibrahim *et al.*, 2008). Data show that the spider mite population increased as dense of hairs increased and vice versa.

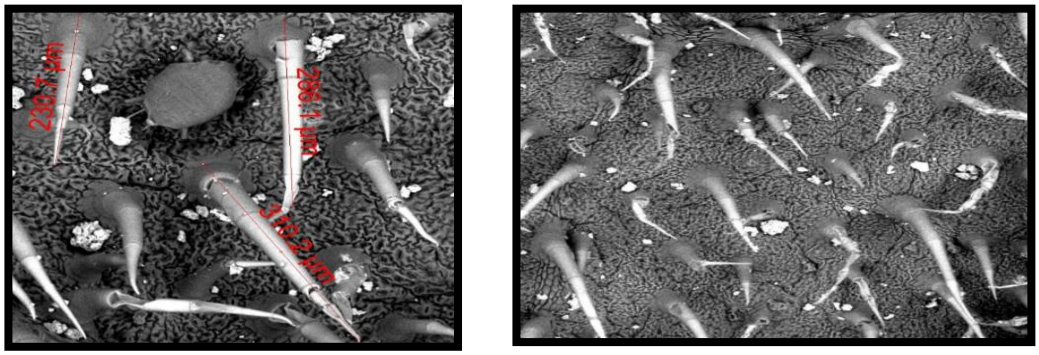
Table (4) : Measurements of some morphological characters of squash leaves in three cultivars.

Cultivar	Density of trichomesin 1mm	Length of smallest trichome in µm			Length of longest trichome in µm		
		length with base	length without base	size base	length with base	length without base	size base
Mabroka	29	106.25	50	56.25	244.7	174.6	70.1
Brencessa	39	112.5	56.25	56.25	310.2	285.2	25
Eskandarani	61	106.25	37.5	68.75	309.3	267.4	41.9

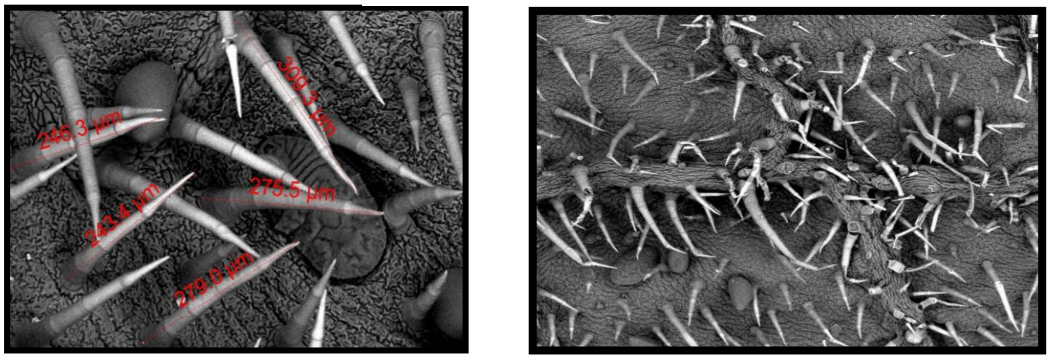
The newly developed setae with base size are 56.25, 56.25 and 68.75 µm for Mabroka, Brencessa and Eskandarani, respectively (Table 4). Base of old trichome of Mabroka cv. increased in size and became 70.1 µm and decreased in Eskandarani cv. became 41.9 µm, while in Brencessa cv., the base diminished more than half recorded 25 µm. The obtained data indicated that there were correlations between the phytochemical contents and the rate of mite infestation.



Mabroka



Brencessa



Eskandarani

Figure (5): Scanning electron micrograph of lower surface leaves showing trichomes.

There is a positive relationship between the infestation levels and the total sugar contents in the 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 results confined that the increasing of total sugar content lead to an increase in the population of phytophagous mite species on the tested cultivars, while a negative relationship was detected with total phenol and free amino acids. 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. 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 the Mabroka cultivar was the most resistant cultivar to the spider mite pest as its leaves harbored the lowest mean numbers and produced higher average of yield than that of Eskandarani cultivar in both villages, followed by Brencessa cultivar which occupied a moderate mean numbers of the spider mite in spite of produced higher total of yield production than that of Eskandarani cultivar. Which suffered the highest infestation of the spider mite population during both seasons so its production of fruit yield was very weak.

It could be classified to three degrees, the severe infestation as represented by (Eskandarani cv.), the moderate infestation (Brencessa cv.) and the last of low infestation (Mabroka cv.). It is concluded that growing the Mabroka cultivar better than the other two tested cultivars.

Conflict of Interest

The present study was performed in absence of any conflict of interest.

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