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Relative Abundance of the Silverleaf Whitefly, *Bemisia Argentifolii* Bellows and Perring, on Squash Plants in Egypt

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Abstract: Two biotypes of the whitefly, *Bemisia tabaci*, were recorded in Dakahelia and Damietta Governorates, Egypt. Biotype "B" or *Bemisia argentifolii* has a wide host range occurring on squash, tomato, cabbage, cotton and duranta plants, while Biotype "Q" was recorded only on lantana plants. Squash plants attracted the silverleaf whitefly (SLWF), *B. argentifolii*, in both spring and fall plantations with significantly higher abundance in the latter. The correlation coefficient between SLWF life stages was discussed. Occurrence of the SLWF on squash plants indicated that the middle and lower leaves were preferred to nymphs and adults. Meanwhile all squash leaves were suitable for SLWF females to lay eggs, with higher deposition on the middle ones. In the field, *B. argentifolii* infestations were associated with silvering leaves disorder. The symptoms appeared after 21.5 and 17.5 days in the spring and fall plantations, respectively. Meanwhile in the laboratory, the same symptoms were observed 16.5 days after infestation.

Key words: Ecological studies, relative abundance, whiteflies, *Bemisia tabaci*, *Bemisia argentifolii*, biotypes A, B, and Q, squash silverleaf disorders, within plant distribution.

Introduction

Since 1986, the population of *Bemisia* species complex has increased to the epidemic levels. In last decade, the genus *Bemisia* received attention of Entomologists and growers throughout the world. In the USA, *Bemisia* spp. caused serious losses in agricultural production exceeding \$ 200 millions in 1991 with an additional annual loss off between 3000-6000 jobs (Henneberry *et al.*, 1993). In Egypt, it has been also emphasized that the populations of *Bemisia* spp. have recently risen to become serious pests attacking all crops from 1990 till now (Tantawi, 1995; Khodeir, 1997; Abdel-Baky *et al.*, 1998; El-Sayed *et al.*, 1998 and Samy, 1999).

Based on their genomic and behavioral studies, Perring *et al.* (1993) identified two types or strains of *Bemisia tabaci* biotype "A" (cotton strain, or *B. tabaci* strain- A) and the other is called biotype "B" (poinsettia strain, or *B. tabaci* strain - B). Bellows *et al.* (1994) described the biotype "B" as a new species, *Bemisia argentifolii* according to the morphological and allozymic characters.

Certain biological features are used to distinguish between the two species *B. argentifolii* (biotype "B") is smaller in size and lays significantly more eggs than *B. tabaci*, biotype "A" (Bethke *et al.* 1991). The feeding rates of biotype "B" are higher, sucking greater quantities of plant sap and producing massive volumes of honeydew (Byrne and Miller, 1990). Moreover, *B. argentifolii* has the ability to induce physiological disorders such as the silverleaf of squash (Schuster *et al.*, 1990; Secker *et al.*, 1998), irregular ripening of tomato (Powell *et al.*, 1998), light root in carrots and light stalk in broccoli (Brown *et al.*, 1991).

Recent outbreaks of *Bemisia* spp. on different crop hosts in the world agro-ecosystem may result from the introduction of the new species, *B. argentifolii* Bellows & Perring, which is thought to be more virulent species (Perring *et al.*, 1993; Bellows *et al.*, 1994). The difficulty of controlling this species is due to many reasons: its widespread distribution on a wide host range (Cock, 1986), short generation time (Butler *et al.*, 1986) and its resistance to, approximately, all insecticide groups (Prabhaker *et al.*, 1996).

The most severe problems in the vegetable production on the worldwide scale arise from the ability of the silverleaf whitefly (SLWF) to transmit more and new geminiviruses diseases to many host plants (Secker *et al.*, 1998). Vegetable crops infested with SLWF risk rejection on both domestic and international markets and even the presence of symptoms of the insect infestations is considered unacceptable. These marketing realities exacerbate the impact of *B. argentifolii* on squash and tomatoes plants. In Egypt, the symptoms of silverleaf disorders in squash, as a sign

of the infestation with the biotype "B", *B. argentifolii*, were observed at different localities such as Delta, Giza, El-Faiyum and Aswan (Lacey *et al.*, 1993; Abdel-Baky *et al.*, 1998). The esterase electromorph analysis on the Egyptian whiteflies collected from tomato fields proved the appearance of the strain "B" in Egypt (Brown *et al.*, 1995). Nevertheless, no additional information exist on the presence of this new species, distribution, population density on another host plants in Egypt (Tantawi, 1995; Khodeir, 1997; Abd El-Kareim, 1998; El-Sayed *et al.*, 1998; Samy, 1999). However, the main objectives of the present investigation are: 1) to identify the *Bemisia* species that infest squash, tomatoes, cotton, duranta, cabbage and lantana in Dakahelia and Damietta Governorates; 2) to determine the abundance of all life on squash plantations; 3) to study the vertical distribution of this species on the leaves of the squash plants; and finally 4) to refer to the weather factors governing the insect distribution.

Materials and Methods

Whitefly Identification: Samples of whiteflies adults were collected by using an aspirator during the morning (from 06 -10 am) from different host plants and kept in Ethanol 90 per cent. These samples were sent to Dr. Pual de Barro Lab., CSIRO Entomology, Canberra, Australia for identification by using Randomly Amplified Polymorphic DNA (RAPD-PCR) technique.

Plant host: This research was conducted at the Experimental Research Station (ERS) of Mansoura University inside the campus. The area of squash plantation per each season was about half feddan.

Since the squash plants are very sensitive to *Bemisia* spp. infestations, therefore this crop was chosen to these studies. The plants were cultivated twice yearly, one during spring and the other during the fall for the two successive seasons 1996 & 1997. The squash plants received the normal agronomic practices during the growing season.

Whitefly Sampling: Because of the difference of behavior of the whitefly stages, two sampling procedures were employed to evaluate the population density of this insect as follows:

Whitefly Adults Sampling Trap: Yellow sticky traps (7.6 x 7.6 cm card) were placed horizontally 15 cm above the ground at 20 m distances along the squash fields. The traps were distributed to cover all the field area. The traps were collected weekly and

replaced by new ones. Counting of *Bemisia* adults was fulfilled by dividing the traps to squares centimeters and calculating the total of adults per trap card.

Sampling of *Bemisia* immature stages: Leaf sampling was started 15 days after germination. Fifteen plants were selected randomly each week, three leaves from each plant were taken, one from upper, middle, and the third from old leaves (lower). The leaves from each category were put in a plastic bag and transferred to the laboratory. According to Abisgold and Fishpool (1990), the leaf of squash plant was divided to three sectors, two involving the edge of leaf and the other in the middle. Two squares inches were measured inside each sector and number of eggs and nymphs were recorded with the aid of a stereomicroscope. The average number of eggs and nymphs per leaf was also calculated.

Vertical distribution: To study the vertical distribution of different stages of *B. argentifolii*, ten plants were chosen and marked for adults study. The squash plant was divided to three categories. The first represented the three top leaves (newly expanded leaves). The second category included leaves located on the fourth and fifth nodes (middle of squash plant). The third involved the lower leaves (from number 6- 8 or 9). Numbers of SLWF adults were recorded visually early in the morning (6-8 a.m.) once a week during the growing season, using the leaf turn method. Considering the nymphs and eggs, five squash plants were chosen randomly each week. Leaves from each category were cut off, placed in the plastic bags and the numbers were recorded by the aid of a stereomicroscope in the laboratory.

Squash Silverleaf (SSL) assessment:

Field trials: In the field studies, the number of silvery leaves of squash, due to the feeding of *Bemisia* spp. (biotype "B") was followed and recorded. The degree of silvery symptoms on the leaves was expressed as rate and classified to four scales: 0 = asymptomatic; 1 = slight silvering; 2 = moderate silvering and 3 = heavy silvering (Cohen *et al.*, 1992). The relationship between *Bemisia* infestation and degree of silvery leaves was estimated.

Laboratory trials: Ten pots of 20-cms in diameter were cultivated with the same variety of squash. With the appearance of the first true leaf, unknown numbers of "B" biotype adults, collected from squash field, were introduced to the plants and maintained for three days under screen cages, and then released. The symptoms of the silverleaf were studied.

Statistical Analysis: All experimental data concerning the above characters were analyzed with one way analysis of variance (ANOVA). Comparisons of biological characters were made according to the Duncan's Multiple Range Test (Costat Software, 1990).

Results

Whitefly Identification: The data in Table 1 show the results of RAPD-PCR analysis of the Egyptian whiteflies collected from different plant hosts in Lower Egypt. It may be obvious that all whiteflies collected from squash, tomato, cabbage, cotton and duranta were silverleaf "B" biotype, *Bemisia argentifolii* Bellows and Perring. Meanwhile, the whiteflies collected from lantana plants in Dakahelia were the non-silverleaf whitefly, which are relatively close to the biotype "B".

Population Dynamics of *B. argentifolii* on Squash Plants:

Adults: Absolute numbers of SLWF adults differed in the two experimental years, although their seasonal distribution was remarkably similar (Table 2). In both years, the adults of *B.*

argentifolii were attracted to squash plants immediately after germination. Table 2 indicates the average numbers of various stages of SLWF adults during 1996 and 1997. The mean numbers of adults per yellow sticky cards (YST) were 360.78 ± 82.49 and 355.16 ± 53.62 in spring plants. In the fall, the corresponding average numbers were 524.36 ± 103.2 and 484.11 ± 97.0 adults in 1996 and 1997, respectively. The percentage of trap catches of the adults formed 58.1 per cent in the fall (27.71 and 30.39% in 1996 & 1997, in succession) and 41.90 per cent in the spring (20.96 and 20.94% in 1996 & 1997). Ideally, the data revealed that the average numbers of *B. argentifolii* varied significantly according to the growing season. SLWF numbers were higher in the fall plantations than in the spring plantations (Table 2) during the two successive seasons.

Fig. 1 also illustrates the populations of SLWF adults which fluctuated during the season. The population densities of *B. argentifolii* adults rose slightly at first in squash fields and then increased dramatically till the end of the season, reaching levels about 531.7 and 440.1 adults per card in the spring. In the fall, there were 667.8 and 603.1 adults per yellow sticky card in 1996 & 1997, respectively.

Eggs: The average number of SLWF eggs was high on squash plants during 1996 & 1997 (Table 2). The average number of eggs were 43.76 ± 4.34 and 46.37 ± 9.83 /inch² in spring plantations and 79.89 ± 16.11 and 79.25 ± 22.17 per inch² in fall during 1996 and 1997, successively. The numbers of SLWF eggs laid on squash plants constituted about 64.16 per cent and 35.84 per cent in the two seasons of fall and spring, respectively. This means that the numbers of eggs laid in the fall was almost twice as much as that laid in spring plantations. The mean numbers of eggs varied significantly in fall than in spring ($P=0.05$). In addition, Figure (1) shows that the average number of eggs was low at the beginning of infestation and then increased to higher levels towards the end of the season.

Nymphs: It may be clear in Table 2 that the mean numbers of nymphs per inch² were 11.17 ± 2.92 and 11.47 ± 2.94 during spring in both 1996 and 1997, respectively. Meanwhile in the fall these averages were 13.52 ± 2.71 and 15.27 ± 2.99 nymphs per inch², successively. However, the numbers of nymphs formed about 56.5 per cent and 43.5 per cent in the fall and spring seasons. Generally, the number of nymphs rose by 1.5 to 2 times at the end of season (Fig. 1). The statistical analysis showed that the number of nymphs did not differ significantly between both seasons ($P=0.05$).

Relationship between SLWF life stage during the squash growing season: Table 3 presents the relationship between different stages of SLWF in squash fields. The records indicate that the differences between the total numbers of SLWF adults and the total number of eggs differed significantly during fall 1996 only ($r=0.7968$) and spring 1997 ($r=0.5635$). Similarly, the correlation coefficient between the total number of SLWF adults and the total number of nymphs were also highly significant ($r=0.7740$) during the fall of 1996 only. Meanwhile, the differences between the total number of eggs and the total numbers of nymphs during the four seasons showed high correlation coefficients. The r values were 0.7029 and 0.9041 during spring of 1996 and 1997, respectively, while they were 0.7026 and 0.9100 in fall of both years 1996 and 1997, in succession (Table 3).

Distribution of various stages on squash plant: The silverleaf whitefly stages differed in their vertical distribution on the squash plant

Abdel-Baky and Abdel-Salam: Relative abundance of the silverleaf whitefly

Table 1: Identification of *Bemisia spp* collected from different plant hosts in Dakahelia and Damietta governorates, Egypt.

Plant hosts	Location	<i>Bemisia spp.</i>
Squash, <i>Cucurbita pepo</i> L	Dakahelia and Damietta	Silverleaf whitefly, biotype "B", <i>B. argentifolii</i> Bellows & perring.
Tomato, <i>Lycoperscum</i> spp.	Dakahelia and Dameitta	Silverleaf whitefly, biotype "B", <i>B. argentifolii</i> Bellows & perring.
Cabbage, <i>Brassica oleracea</i> var. <i>capitata</i> L.	Dakahelia and Dameitta	Silverleaf whitefly, biotype "B", <i>B. argentifolii</i> Bellows & perring.
Cotton, <i>Gossybbium barabadense</i> L	Dakahelia	Silverleaf whitefly, biotype "B", <i>B. argentifolii</i> Bellows & perring.
Duranta, <i>Duranta plumeri</i> var. <i>variegata</i> L.	Mansoura region, Dakahelia	Silverleaf whitefly, biotype "B", <i>B. argentifolii</i> Bellows & perring.
Lantana, <i>Lantana camara</i> L.	Mansoura region, Dakahelia	<i>Bemisia tabaci</i> , biotype "Q".

Table 2: The average numbers of different stages of *Bemisia argentifolii* on squash plants.

Season	Mean numbers ± sx of	Mean numbers ± sx of	Mean numbers ± sx of
	SLWF adults per card	SLWF eggs per inch ²	SLWF nymphs per inch ²
Spring 1996	360.78 ± 82.49 b A	43.76 ± 4.34 b B	11.17 ± 2.92 c C
Fall 1996	524.36 ± 103.2 a A	83.28 ± 16.11 a B	13.52 ± 2.71 ab C
Spring 1997	355.16 ± 53.62 b A	46.37 ± 9.83 b B	11.47 ± 2.94 bc C
Fall 1997	484.11 ± 97.0 a A	79.25 ± 22.17 a B	15.27 ± 2.99 a C

*Means followed by the same letter in a column are not significantly differences at the 5 per cent level of probability (Duncan's Multiple Range Test).

*Means followed by the same letter in a row are not significantly differences at the 5 per cent level of probability (Duncan's Multiple Range Test).

Table 3: Correlation coefficients between different stages of *B. argentifolii* in 1996 and 1997 on squash plants.

Seasons	Relationships between different stages		Correlation Coefficient parameters			
			R	Slope (b)	Y Int (a)	Significant Sign
Spring 1996	Adults	Eggs	0.1990	0.03159	1119.920	NS
	Adults	Nymphs	0.0982	0.01052	29.787	NS
	Eggs	Nymphs	0.7029	1.04106	96.329	**
Spring 1997	Adults	Eggs	0.5635	0.3097	15.705	*
	Adults	Nymphs	0.3161	0.0521	27.8144	NS
	Eggs	Nymphs	0.9041	3.0159	35.290	***
Fall 1996	Adults	Eggs	0.7968	0.3574	55.591	**
	Adults	Nymphs	0.7740	0.0583	10.500	**
	Eggs	Nymphs	0.7026	4.1831	70.1134	*
Fall 1997	Adults	Eggs	0.1217	0.0835	197.343	NS
	Adults	Nymphs	0.4351	0.0402	26.322	NS
	Eggs	Nymphs	0.91003	6.7485	-71.326	***

Table 4: Distribution of various stages of *B. argentifolii* on squash plant.

Seasons	% of SLWF eggs			% of SLWF nymphs			% of SLWF adults		
	Top	Middle	Lower	Top	Middle	Lower	Top	Middle	Lower
Spring 1996	31.36 b	34.55 a	34.09 a	00.00 c	45.34 b	54.66 a	10.60 b	21.45 b	67.95 a
Spring 1997	30.56 b	36.59 a	32.85 b	00.00 c	45.85 b	54.15 a	08.26 b	36.35 b	55.39 a
Fall 1996	30.19 b	36.89 a	32.92 b	00.00 c	45.27 b	54.73 a	15.20 b	31.05 b	53.75 a
Fall 1997	28.74 c	38.55 a	32.17 b	00.00 c	48.12 b	51.88 a	13.77 b	30.47 b	55.76 a

*Means followed by the same letter in a row are not significantly differences at the 5 per cent level of probability (Duncan's Multiple Range Test).

Table 5: Correlation coefficients of the distribution of the stages of *B. argentifolii* on squash plants during 1996 & 1997.

Season	SLWF stages relationships on leaf position.		Correlation Coefficients			
			R	Slope (b)	Y Int (a)	Significant Sign
Spring 1996	Nymphs on middle leaves	Nymphs on lower leaves	0.9408	1.0781	1.9369	***
Spring 1997	Nymphs on middle leaves	Nymphs on lower leaves	0.9555	1.0808	1.54717	***
Fall 1996	Nymphs on middle leaves	Nymphs on lower leaves	0.9669	1.1350	1.4095	***
Fall 1997	Nymphs on middle leaves	Nymphs on lower leaves	0.9846	0.9994	1.7325	***
Spring 1996	Eggs on middle leaves	Eggs on upper leaves	0.9168	1.0179	-8.6540	***
	Eggs on middle leaves	Eggs on lower leaves	0.9247	0.8349	3.5453	***
Spring 1997	Eggs on middle leaves	Eggs on upper leaves	0.9337	0.7722	4.4002	***
	Eggs on middle leaves	Eggs on lower leaves	0.9227	0.9166	0.4469	***
Fall 1996	Eggs on middle leaves	Eggs on upper leaves	0.8976	0.6032	19.020	***
	Eggs on middle leaves	Eggs on lower leaves	0.9800	0.8101	7.2291	***
Fall 1997	Eggs on middle leaves	Eggs on upper leaves	0.9673	0.8027	4.1970	***
	Eggs on middle leaves	Eggs on lower leaves	0.9839	2.4266	15.3657	***

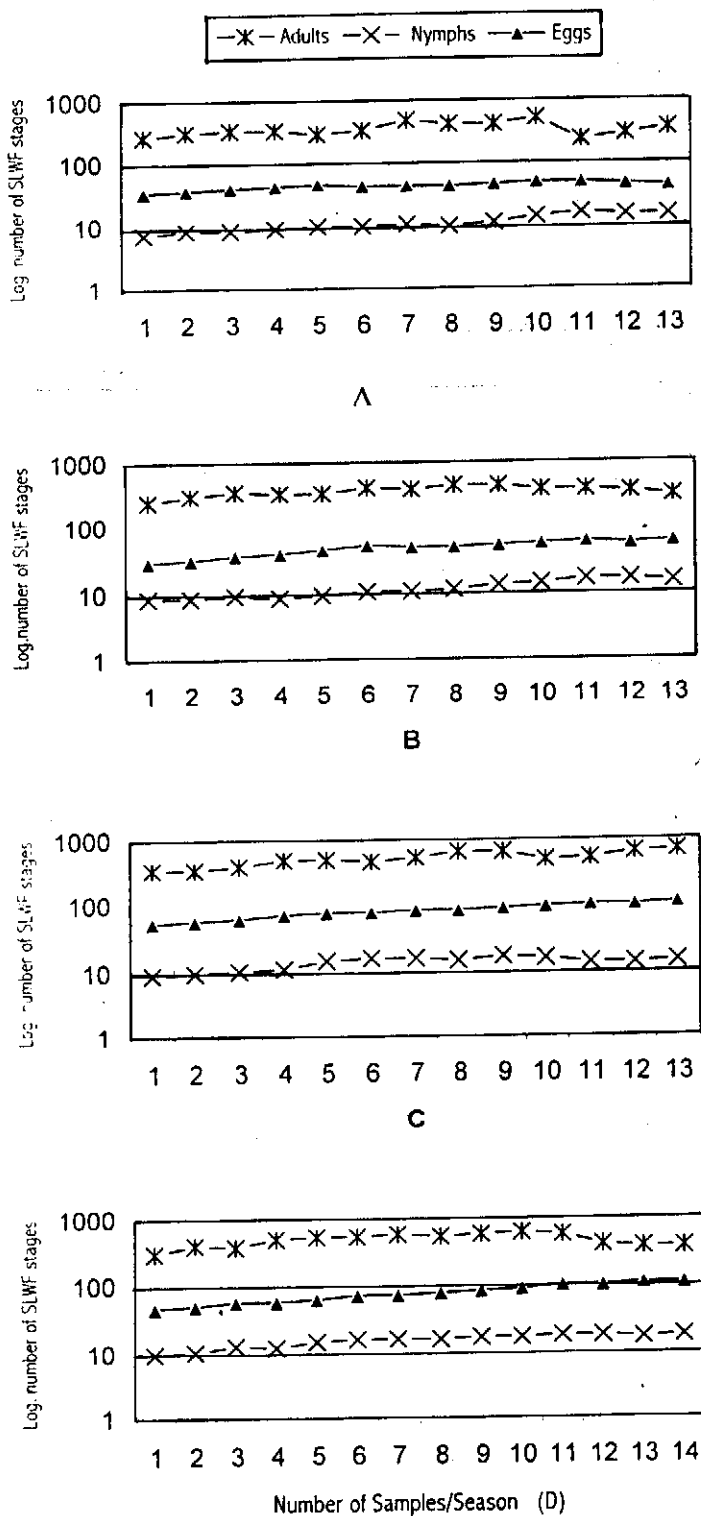


Fig. (1): Log number of *B. argentifolii* life stages on squash plants during 1996 & 1997.
 A): Spring 96 B): Spring 97 C): Fall 96
 D): Fall 1997

(Table 4). They occurred on all leaves of the plant except the nymphs which were not present on the top leaves. As far as eggs, greater numbers were found the middle leaves (fully expanded leaves) of squash (from node 4-5) which formed about (34.55-38.55%) of the total fecundity, followed by the lower leaves (32.71-34.09%). Meanwhile, the top leaves had a low percent of the eggs (28.74-31.36%). The statistical analysis proved that there were significant differences between the leaves considering SLWF eggs distribution ($P=0.05$).

The distribution of *B. argentifolii* nymphs on squash plant was limited only on both middle and lower leaves (Table 4). The nymphs constituted from 45.27 to 48.12 per cent of their population. This percentage was higher on lower leaves being 51.88 - 54.73 per cent. The statistical analysis of variance clarified that there were significant differences ($P=0.05$) between the numbers of nymphs on the squash leaves. It was observed that the SLWF nymphs preferred the lower leaves, particularly the red-eyed nymphs. It is also evident in Table 4 that adults formed 8.26 - 15.2 per cent on the top leaves, 21.45-36.35 per cent on the middle leaves and 53.75 - 67.95 per cent on the lower ones. In addition, data showed that the adult distribution had a great significant variance on squash plant according to leaf age and position on the main stem of the plants.

Table 5 presents the correlation coefficients between the insect stages and their vertical distribution on squash plant. It may be obvious that the correlation between the average numbers of SLWF nymphs on both the middle and lower leaves was highly significant. Nevertheless, the correlation coefficient between the average numbers of nymphs on both the top, middle and lower leaves, was infinity. In addition, the correlation coefficients between the numbers of eggs on the previous levels of leaf distribution were significant with higher values of "r" which reached above 0.9.

Correlation coefficient between the stages of *B. argentifolii* and certain weather factors: Table (6) gives the relationship between the average numbers of SLWF adults, nymphs and eggs and four weather factors that influenced the development, migration, feeding process and damage caused by the insect.

Regarding the correlation between the number of laid eggs and temperature was significant in both spring and fall of 1996 and 1997. The "r" values of both minimum and maximum temperature were 0.686, 0.683 and 0.908, 0.823 in spring of 1996 and 1997, while in the fall, the corresponding values were 0.971, 0.983 and 0.931, 0.919 in both years. Moreover, relative humidity (R.H.) showed insignificant effects between the number of the deposited eggs, particularly with the maximum R. H. in the four seasons and minimum R. H. in spring of 1996 only (Table 6).

The effect of temperature on the number of SLWF nymphs were significant during 1996 & 1997 (Table 6), where "r" values of minimum temperature were 0.834, 0.806 in the spring and -0.802, -0.904 in the fall plantations during 1996 & 1997, respectively. Ideally, the values of "r" for maximum temperature were 0.692, 0.597 and -0.732, -0.740 in both spring and fall seasons during 1996 & 1997. In addition, the data also show that the effect of temperature was significantly negative in fall plantations only. Both minimum and maximum R. H. clarified a non correlation coefficient between the numbers of SLWF nymphs (Table 6), except the minimum R. H. during spring and fall 1997. As far as adults, no significant effects of both temperature and relative humidity on their population in 1996. Nevertheless, the minimum and maximum temperature and minimum R. H. affected the fall population in 1996 and 1997 with great correlation coefficient with the total numbers of the insect.

Squash Silverleaf symptoms and its relationship to *B. argentifolii* infestation: In the field trial, the silverleaf symptoms were observed after 18-25 (with an average 21.5 days) and 16-19 days (with an

Abdel-Baky and Abdel-Salam: Relative abundance of the silverleaf whitefly

Table 6: Correlation coefficient between *Bemisia argentifolii* life stages and certain weather factors in squash fields during 1996 & 1997.

	SLWF life stages											
	Eggs				Nymphs				Adults			
	Temperature		R. H.		Temperature		R. H.		Temperature		R. H.	
	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.
1996												
Spring												
r	0.686	0.683	0.165	0.1398	0.834	0.692	0.537	-0.283	0.302	0.261	-0.801	-0.042
Slope B	0.140	0.187	0.060	0.038	0.253	0.281	0.287	0.114	9.300	0.012	-0.005	-0.002
Y Int (a)	-0.548	7.338	24.34	85.72	9.39	22.48	22.53	94.50	193.5	27.83	33.82	91.31
Signi.	**	*	NS	NS	***	**	NS	NS	NS	NS	NS	NS
Fall												
r	-0.971	-0.983	-0.849	0.344	-0.802	-0.732	0.536	0.325	-0.847	-0.814	-0.724	0.732
Slope B	-0.088	-0.101	-0.050	0.017	-0.435	-0.448	-0.189	0.101	-0.035	-0.038	-0.019	0.008
Y Int (a)	38.89	54.62	44.71	84.77	35.31	48.54	45.37	84.65	35.51	49.73	47.62	84.60
Signi.	***	***	***	NS	**	**	NS	NS	***	**	**	NS
1997												
Spring												
r	0.908	0.823	0.691	-0.368	0.806	0.597	0.842	-0.339	0.563	0.753	0.003	0.336
Slope B	0.128	0.075	0.180	-0.069	0.379	0.182	0.733	-0.042	0.044	0.038	3.250	-0.007
Y Int (a)	-0.352	22.47	4.123	88.04	4.402	26.66	4.001	87.58	1.781	19.35	29.12	88.61
Signi.	***	***	**	NS	***	*	***	NS	*	**	NS	NS
Fall												
r	-0.931	-0.919	-0.595	-0.505	-0.904	-0.740	-0.779	-0.442	-0.181	0.151	-0.536	0.026
Slope B	-0.041	-0.036	-0.424	-0.005	-0.291	-0.214	-0.412	-0.030	-0.005	0.004	-0.026	1.549
Y Int (a)	26.35	34.29	46.55	86.42	30.06	40.56	55.33	86.69	19.36	28.84	49.14	85.26
Signi.	***	***	*	NS	***	**	**	NS	NS	NS	*	NS

Table 7: Average numbers of squash silvering leaves (SSL) and regression analysis between SLWF nymphs and silvering leaves.

Seasons	Average numbers of Silvering leaves			Multiple regression equations
	Top	Middle	Lower	
Spring 1996	1.136 b	1.815 ab	2.154 a	Y = 6.326 + 0.949x1 + 1.529 x2 (r ² = 0.6601).
Spring 1997	1.175 b	1.764 ab	2.193 a	Y = 7.076 + 1.435x1 + 0.869x2 (r ² = 0.6873).
Fall 1996	1.683 b	2.339 ab	2.662 a	Y = 8.427 - 3.907x1 + 5.125 x2 (r ² = 0.7140)
Fall 1997	1.569 b	2.068 ab	2.536 a	Y = 8.568 - 0.437 x1 + 2.707 x2 (r ² = 0.9393)

*Means followed by the same letter in a row are not significantly differences at the 5 per cent level of probability (Duncan's Multiple Range Test). x1 & x2 means the number of silverleaf on both middle and lower position of main stem.

average 17.5 days) during the spring and fall plantations, respectively. The symptoms were observed on all ages of squash leaves. Some non-silver symptoms appeared only on the leaves

located on node one of squash stem (Scale 0 = asymptomatic). A slight degree of symptoms was recorded on the leaves from node 2-3 from the top (scale 1). Meanwhile moderate degrees of silver symptoms were noticed on the leaves 4 and 5 (scale 2), and scale 3 which has a heavy degree of squash silvering symptoms on lower leaves (from 6 to oldest one).

In the laboratory trials, the silver symptoms or/vein clearing appeared 16.5 days after SLWF infestations. The evidence of laboratory studies indicated that the vein clearing began as chlorosis around the main veins of the youngest leaves (grade 1), then increased gradually to include the other veins of the leaf (grade 2). With the progress of developing infestation and the period of infestation, all leaf area between veins was colored with silver symptoms (grade 3).

The statistical analysis of variance revealed that the relation between number of silver leaves on the previous scales was significant (Table 7). In addition, the multiple regression analysis between average numbers of *B. argentifolii* nymphs and the average

numbers of vein clearing leaves was highly significant. The equations and r² values were Y = 6.326 + 0.949x1 + 1.529 x2 (r² = 0.6601), Y = 7.076 + 1.435x1 + 0.869x2 (r² = 0.6873), Y = 8.427 - 3.907x1 + 5.125 x2 (r² = 0.7140) and Y = 8.568 - 0.437 x1 + 2.707 x2 (r² = 0.9393) for spring and fall 1996 & 1997 (Table 7), respectively.

Discussion

In recent years, *Bemisia* spp. have become the most serious pests of the family Aleyrodidae attacking over 500 plant hosts (Cock, 1986; Secker *et al.*, 1998). The identification of various species is highly important, as it helps to study the biological characters, to understand the behavior of the insect, to minimize its harmful effects and to plan a suitable control strategy on ecological bases. Through crossing experiments, mating compatibility studies and analysis of genetic differences, Perring *et al.* (1993), differentiated two biotypes of *Bemisia* namely, *Bemisia tabaci* biotype "A" (cotton-strain) and *Bemisia tabaci* biotype "B" (poinsettia-strain). Moreover, Bellows *et al.* (1994) described the new biotype "B" as a distinct species namely silverleaf whitefly (SLWF) or *Bemisia argentifolii* Bellows and Perring. The RAPD-PCR analysis fulfilled at

CSIRO-Australia on the Egyptian whitefly samples indicated that the new biotype of *Bemisia* (*B. argentifolii* or biotype "B") was recorded in Egypt (Table 1) on many host plants as the most dominant species in Egyptian fields (Abdel-Baky & Abdel-Salam, unpublished Data). These evidences are in an agreement with electromorphs analysis of Brown *et al.* (1995) on the Egyptian whiteflies collected from tomato fields and observations of Lacey *et al.* (1993) and Abdel-Baky *et al.* (1998) in squash fields. The new species, *B. argentifolii* occurs on other plant hosts in Egypt. Presently, the biotype "B" has become well established on both field and greenhouse crops throughout the world (Bedford *et al.*, 1994). There are also many reports emphasizing that *B. argentifolii* is worldwide (Lacey *et al.*, 1993; Brown *et al.*, 1995; Gunning *et al.*, 1995; Guirao *et al.*, 1997; Rosell *et al.*, 1997; Berdiales *et al.*, 1999). The reasons that make SLWF such a concern is its tendency to rapid development to an extremely high populations (Bethke *et al.*, 1991), its broad host ranges (Cock, 1986; Secker *et al.*, 1998), together with its association with some viral diseases (Brown and Bird, 1995).

The present investigation of the Egyptian whiteflies proved the existence of another biotype of *Bemisia tabaci*, known as biotype "Q". Rosell *et al.* (1997) reported clearly that *Bemisia tabaci* has other biotypes. These biotypes are non-A/ non-B and others marked K, L, P and Q. The biotype "Q" found only on Lantana plants in Egypt (Table 1), is a non-silvering and is a serious pest in Spain, Sudan (De Barro, personal communication, 1999) and Portugal (Berdiales *et al.*, 1999) as a close relative of the "B" biotype.

The two biotypes of *Bemisia* (B & Q) exhibit a similar efficiency in transmission cucurbit yellow stunting disorder virus (CYSDV) under controlled condition (Guirao *et al.*, 1997). In contrast, the biotype "A" (*Bemisia tabaci*) is an inefficient vector of CYSDV when compared with biotype "B" (Wisler *et al.*, 1998).

Regarding SLWF origin and distribution, Wool *et al.* (1994) suggested that the origin of biotype "B" may be Central and South America and spread to the Caribbean and the USA. While, Bedford (at UK) proposed that the Middle East may be the origin of *B. argentifolii* (De Barro 1995) due to the earliest records of the silverleaf symptoms in Israel (Paris *et al.*, 1987).

The data in Table 2 indicate that the squash plants were attractive and suitable for SLWF feeding, oviposition and development. SLWF population densities were smaller in the spring than in the fall plantations (Fig. 1). In general, the population fluctuations of SLWF cycled independently from field to another. However, a pattern emerged where numbers of SLWF increased steadily through late summer with the peak densities from the beginning of fall. This may be due to the harvest dates of the summer crops (Legaspi and Carruthers, 1995). Similarly, Godfrey *et al.* (1994) pointed out that the numbers of SLWF tended to build up slowly in the spring before increasing exponentially through the summer and early fall.

To develop a better sampling program, it's necessary to understand the distribution of SLWF population on the plant (Naranjo and Filint, 1994 and 1995). The distribution of *B. argentifolii* on an individual plant is mostly a function of the interaction between the oviposition behavior of the females and sessile habit of the immature stages with squash growth characteristics (Tonhasca *et al.*, 1994). The squash leaf age and height is an important factor influencing *B. argentifolii* distribution on squash plant. The present results indicated that young leaves at the middle height of the plant were preferred as oviposition site than the youngest ones at the upper main stem and those on the lower height of the plant. These results are closely similar to those of Gameel (1977); Ohnesorge *et al.* (1980); Liu and Stansly (1995). Their results showed that the whitefly females deposited more eggs on young leaves at the middle than other leaves of the plant.

The vertical distribution of SLWF nymphs occurred only on both middle and lower leaves. The third nymphal instars and pupa were

concentrated on the older leaves only, while newly hatched nymphs and the second instar aggregated on the middle leaves. This may be due to the growth of the plant (Ohnesorge *et al.*, 1980; Naranjo and Filint, 1994; 1995). In addition, the adults occupied with higher percentage on lower leaves (53%). The behavior of the adults is apparently responsible for the vertical distribution on the plant. The emergence of adults occurred on the lower leaves in the morning (Butler *et al.*, 1986). Therefore, the adults choose canopy leaves for protection and movement between the leaves of the plant for feeding and oviposition (Butler *et al.*, 1986). The females select youngest leaves for oviposition, probe more frequently, spend less time probing and feeding on the upper leaves (Ohnesorge *et al.*, 1980; Noldus *et al.*, 1986; Naranjo and Filint, 1995). *Bemisia argentifolii* vertical distribution is mostly enhanced by the chemical constituents of the leaves (Noldus *et al.*, 1986; Janssen *et al.*, 1989), as well as, by their morphological characteristics (Kamp and Lenteren 1981; Simmons, 1999).

However the results of the insect distribution on the plant were incompatible with that of Lynch and Simmons (1993), who found that the whitefly immature stages were most abundant in peanuts on the leaves 3, 4 and 5. A study of vertical distribution of *Bemisia* of the tomato plants revealed that eggs were laid on newly formed leaves and nymphs were found on leaves 3-7. Red-eyed nymphs were detected on leaf 8 and lower leaves (Ohnesorge *et al.*, 1980). The vertical distribution of *Bemisia tabaci* in the cotton canopy depended on the cotton variety and plant growth stage (Arx *et al.*, 1984).

Bemisia argentifolii is known as a thermophilic pest (Avidov, 1956). It appears to be well-adapted to develop on squash plants, whereas it is able to build up a high population density throughout the year, although it was more abundant in the fall than in spring in the two successive years of study. Since temperature is necessary for the insect development, the life span from egg to adult decreased at higher temperatures (Butler *et al.*, 1983; Wanger, 1995), which may explain the occurrence of SLWF high population densities during late summer and fall. The present result likewise prove that temperature is a major weather factor influencing the development and population size of SLWF more than relative humidity.

Because SLWF eggs are physiologically indirect contact with plant stomata (Pollard, 1955; Byrne *et al.*, 1990), the effects of R.H. are insignificant on embryonic development and eggs hatching, which may be due to the greater occurrence of the whitefly on the lower surfaces of the plant leaves (Coombe, 1982). Horowitz *et al.* (1984) also demonstrated that changes in the whitefly population are attributed mainly to weather factors such as temperature and relative humidity. However, the statistical analysis of the present results indicated that temperature had a greater impact on SLWF population on squash fields than relative humidity (Table 7).

It has been reported by Secker *et al.* (1998) that silverleaf in cucurbit plants reflects disorder associated with *B. argentifolii* infestation. In the present work, squash plants which are more susceptible to silver leaf than other cucurbits, showed limited degrees of symptoms. Similar results were obtained by Schuster *et al.* (1990) who found that the slight and moderate vein blanching occurred on leaves 5 and 6 on the main stem from upper, while the lower leaves were completely silvering. Also, Secker *et al.* (1998) reported that silvering degrees were highly correlated with the numbers of SLWF adults and nymphs. In contrast, Jimenez *et al.* (1995) mentioned that the silvering symptoms were induced by SLWF nymphs feeding only and not due to mechanical effects of stylet contact. The histological basis of leaf vein clearing in squash and other Curcubitaceae is now quite evident. Air spaces beneath the epidermis in the parenchymal cell layer of silvered leaves increase reflection of incident light off the leaf surface (Jimenez *et al.*, 1995). Silverleaf disorder is of economic importance because the affected tissue reduces chlorophyll and high light reflectance

that may result in lower yield (Burrger *et al.*, 1988).

In conclusion, these results show that *Bemisia argentifolii* is an important, most abundant and a serious pests attacking field and vegetable crops in Egypt. Further ecological and physiological studies are required.

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