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Occurrence of major mite species and their biocontrol agents on soybean *Glycine max* crop

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Abstract:

This study conducted to incidence and determined the levels of infestation of phytophagous mites and its predators associated with soybean. There were five species belonging to three families were determined in total collected samples can be classified according to their feeding habitats to two major groups *i.e.* phytophagous mites represented by two species, *Tetranychus urticae* Koch and *Oligonychus pratensis* (Banks) (Acari: Tetranychidae) and predaceous mites represented by three species, *Euseius scutalis* (Athias-Henriot) (Acari: Phytoseiidae), *Agistemus exsertus* Gonzalez (Acari: Stigmaeidae) and *Typhlodromus pyri* Scheuten (Acari: Phytoseiidae) . Frequency of occurrence for mite species associated with soybean were determined. The most distributed species was *T. urticae* followed by *E. scutalis, A. exsertus, T. pyri* while, *O. pratensis* was the lowest species in occurrence. Their percentages of relative occurrence were 34.92, 25.40, 20.63, 11.91 and 7.14%, respectively. *T. urticae* showed two peaks of abundance. The highest peak was 29.25 individual/leaf appeared in the beginning of August month. There were significant differences in population between the infestation % and damage of *T. urticae* during the growth stages of soybean. The percent of protein contents was high through flowering and reproductive stage compared with vegetation stage. Multiplication of *T. urticae* number on soybean, 10, 20 and 30 mites/leaf in greenhouse during growing season, 35 days old soybean plants infested with spider mites at the rate of 30 mites/leaf produced significantly a greater number of spider mites. the age of the soybean plant had no effect on the predator population.

Introduction

Soybean *Glycine max* L. is considered the most important cash crops in many countries and one of the summer legumes crops, with great nutritive value, containing relatively high percentage of oil and proteins many essential amino acids (Badenhop and Hacker, 1971). It is used for feeding human and animals. It can substitute for meat production pross and for some extent for milk and its remains are used as fertilizer to enrich the soil with the steady of increase in world population, the demand for both Soybean oil and protein is still increasing. Many pests infesting soybean crop specially, piercing sucking insects are of the major

insect pests which attack this crop in the fields causing severe damage (El-Kifil *et al*., 1974; Hamed, 1977; Metwally, 1989; Awadallah *et al*., 1991) and El-Khouly *et al*., 1998). *G. max* is subjected to attack by many pests among of these pests, mites throughout the growing seasons. Other mite species may be predators which play an important role in biological control of certain agricultural pests. For example, species belonging to family phytoseiidae, that release of these mites have been shown experimentally to reduce spider mite densities in many crops (Nyrop *et al.*,1998). There is a relationship between both morphology and occurrence of phytoseiid mites and leaf surface texture in addition to weather factors. *Tetranychus urticae* Koch (Acari: Tetranychidae) is the most injurious phytophagous mite on leguminous plants. It feeds on the plant sap causing serious damage according to the rate of infestation. It is also the most abundant species on leaves of vegetable plants in both open field and plastic houses. The population growth of *T. urticae* densities on bean plant at different ages, was studied by Bustos *et al*. (2009) where, bean plants of four weeks of age were an excellent substrate for the development of *T. urticae* population. The present study was carried out to shed light on the mites associated with *G. max*, the study of population dynamics of these phytophagous and predaceous mites during the growing season of *G. max* in addition to the effect of plant age on population and multiplication of *T. urticae* and its predator, *Euseius scutalis* (Athias-Henriot) (Acari: Phytoseiidae).

Materials and methods

1.Occurrence of mite species associated with soybean crop:

Samples of plant leaves were randomly collected every week from soybean *G. max* during growing season 2018. Samples of crop was obtained from district i.e. Zagazig. The total number of collected samples from crop was 123 samples.

1.1.Samples procedures:

Samples were collected in early morning. Each sample consisted of 25 leaves, collected randomly. All collected samples were kept in paper bags. The necessary information including crops, locality and date of collection were recorded. Samples were directly transferred to the laboratory and examined using stereomicroscope. To compare occurrence of the identified species on each crop, percentage of absolute and relative frequency occurrence were calculated according to **Norton (1978)** as follows:

$$\% \text{ Absolute frequency occurrence} = \frac{\text{Number of samples containing species}}{\text{Number of samples collected}} \times 100$$

$$\% \text{ Relative frequency occurrence} = \frac{\text{Frequency of species}}{\text{Sum of frequency of all species}} \times 100$$

1.2.Mounting and identification of surveyed mites:

Collected mite individuals were cleared in Nesbit's solution and mounted in Hoyer's medium on glass slides for identification with the aid of research microscope. The identification according to Krantz (1970).

2.Population dynamics of mites on soybean crop:

Samples were collected in early morning, each one consisted of 25 leaves collected randomly from infested field; all the collected samples were kept in paper bags. Necessary information including crop locality, date of collection and sowing date were directly transferred to the laboratory and examined using stereo microscope to compare occurrence of the identified mite species on the crop,

this process was repeated every week, the recorded temperature and relative humidity associated with the plant in this day was listed. Sample correlation was calculated according to Little and hills (1978). Growing stages of the crop were divided into two stages, vegetation and flowering and reproductive stages to study the effect of plant age on the population of the two spotted spider mite, *T. urticae* to calculate the damage (number of mite individuals/leaf) and the percent of infestation (number of infested leaves/number of total leaves×100).

3. Multiplication of *Tetranychus urticae* on soybean crop:

Soybeans were planted in greenhouses during season 2018 and were divided into 4 treatments according to their age to 25, 35, 45 and 55 days after planting and each treatment included 60 plants were infected with three levels of infection 10, 20 and 30 individuals by 20 plants / level and was taken 5 leaves weekly randomization of each level within each treatment and then counting the mobile individuals.

4. Chemical analysis of soybean leaves:

Soybean leaves were analyzed and estimated, total carbohydrates %, total k (ppm), total p%, total nitrogen %, and total protein %. The leaves were analyzed at agriculture faculty, Benha university, Egypt.

5. Statistical analysis:

Data were subjected to statistical analysis, Duncan (1955), multiple range test was used to determine the significant of the difference between mean values of the treatments.

Results and discussion

Data in Table (1) and Figure (1) showed that five species belonging to three families were determined in total collected samples can be classified according to their feeding habitats to two major groups i.e. phytophagous mites represented by two species, *T. urticae* and *Oligonychus pratensis* (Banks) (Acari: Tetranychidae), and predaceous mites represented by three species, *E. scutalis*, *Agistemus exsertus* Gonzalez (Acari: Stigmaeidae) and *Typhlodromus pyri* Scheuten (Acari:Phytoseiidae).Frequency of occurrence for mite species associated with soybean were determined. The most distributed species was *T. urticae* followed by *E. scutalis, A. exsertus, T. pyri* while, *O. pratensis* was the lowest species in occurrence. Their percentages of relative occurrence were 34.92, 25.40, 20.63, 11.91 and 7.14%, respectively. Species of mite were present with high, moderate and rare abundance. *T. urticae* and *E. scutalis* were most dominant species. These results are similar with Eleawa (2007) reported that the most frequently species was *T. urticae* followed by the predatory mite *E. scutalis* and *Typhlodromis* sp. on soybean plants.

Table (1): Incidence of frequency occurrence (F.O) for mite associated with soybean crop.

Mites	Absolute F. O.%	Relative F. O.%	Frequency
Phytophagous	-	-	-
Family: Tetranychidae	-	-	-
Tetranychus urticae	71.54	34.92	+++
Oligonychus pratensis	14.63	7.14	+
Predaceous	-	-	-
Family: Phytoseiidae	-	-	-
Euseius scutalis	52.03	25.40	+++
Typhlodromus pyri	24.39	11.91	++
Family:Stigmaeidae	-	-	-
Agistemus exsertus	42.27	20.63	++

+++ Highly (10-15) individuals/leaf ++ Moderate (5-10) individuals/leaf + Rare (>2) individuals/leaf

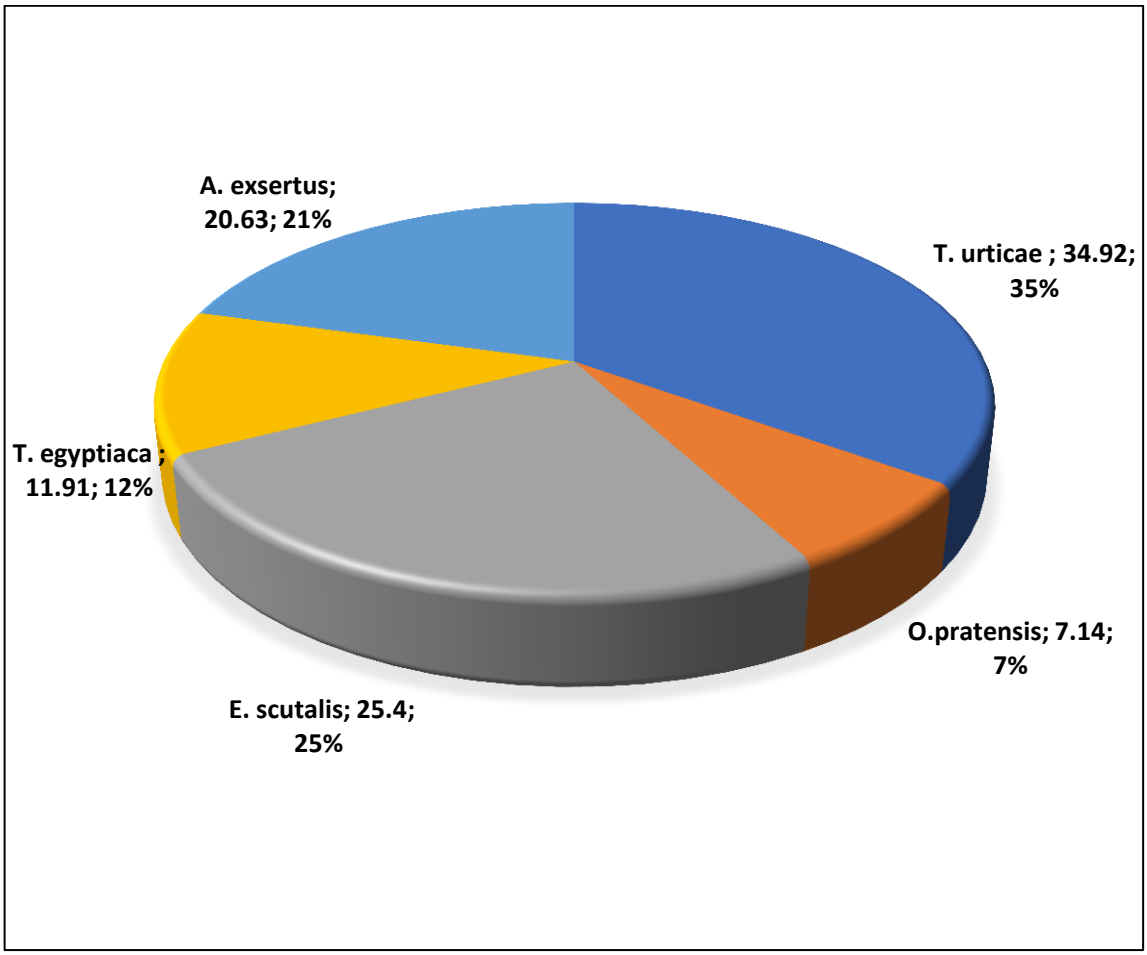

Figure (1): Relative frequency occurrence of mite species associated with soybean crop.

Data in Table (2) illustrated the population dynamics of *T. urticae* on soybean crop during season 2018 weekly the examination was conducted at Zagazig area during fourteen consecutive weeks. Results indicated that *T. urticae* showed two peaks of abundance. The highest peak was 29.25 individual/leaf appeared in the beginning of August month. *O. pratensis* is represented by six peaks the highest peak was 2.53 individual/leaf appeared in 26 May on the other hand in predaceous mite's population fluctuated during the growing season. *E. scutalis* was the most dominant species that appeared with highly population 8 individual/leaf at 11 August while, the population of other predaceous mites appeared with approximate number. The relation between two weather factors temperature (°C) and relative humidity (R53H.) and mite population in Soybean plant indicated significant positive correlation between the population of *T. urticae* and the two weather factors, r=0.72 and 0.39 for temperature and R.H., respectively. while, the correlation was negative between the population of all predaceous mite and R.H. These results are similar with Romeih *et al*. (2013) recorded that the highest peak for adult (26.4, 49.14 and 37.14 individual/leaf for immature and egg, respectively, infestation by *T. urticae* on Faba bean occurred at March.

Table (2): Population dynamics of mites on soybean crop during season 2018.

Date of inspection	Phytophagous mites/leaf		Predaceous mites/leaf		
	Tetranychus urticae	***Oligonychus pratensis***	***Typhlodromus pyri***	***Euseius scutalis***	***Agistemus exsertus***
12/5	0.00	0.23	0.00	0.00	0.00
19/5	2.53	1.50	1.30	0.00	1.50
26/5	7.19	2.53	2.00	0.00	1.00
2/6	10.03	1.37	1.31	1.96	2.30
9/6	15.11	2.04	1.60	2.48	3.20
16/6	12.63	1.56	2.12	3.36	2.24
23/6	5.23	1.80	2.64	4.64	1.60
30/6	5.13	1.32	3.12	5.40	1.28
7/7	9.57	2.24	4.40	6.80	2.80
14/7	14.75	1.16	3.84	4.92	1.96
21/7	18.39	2.16	5.40	4.20	1.80
28/7	21.15	1.68	4.00	5.56	2.80
4/8	29.25	1.20	2.88	7.16	3.32
11/8	21.10	1.96	3.44	8.00	3.96
18/8	13.73	2.44	4.08	7.28	4.20
25/8	5.00	1.52	2.40	3.48	3.20
1/9	2.89	0.88	1.76	2.40	2.88
8/9	2.04	0.76	0.96	1.28	3.20
Mean	10.87	1.57	2.62	3.83	2.40
Correlation coefficient values(r)	**°C**	0.72	0.31	0.66	0.41	0.22
	RH.	0.39	0.45	-0.11	-0.65	-0.28

Data in Table (3) showed that there were significant differences in population between the infestation % and damage of *T. urticae* during the growth stages of Soybean. The infestation% was high 87.27% in flowering and reproductive stage. On the other hand, damage (number of mite/leaf) was high 13.00 individual/leaf in flowering and reproductive stage. These results similar with Anuradha *et al*. (2014) they reported that number of *T. urticae* increased gradually after sowing reached maximum in early growing stage and decreased in delayed growing stage on *Phaseolus vulgaris* L.

Table (3): Damage and infestation % of *Tetranychus urticae* through growing stages of soybean crop during season 2018.

Date of inspection	Growing stage	Infected leaves number/25 leaves	Infection %	Individuals number/leaf (Damage)	Infection %	Damage
12/5	**Vegetative**	00	00	0.00	70.85^{b}	7.53^{b}
19/5		23	92	2.53		
26/5		20	80	7.19		
2/6		20	80	10.03		
9/6		19	76	15.11		
16/6		18	72	12.63		
23/6		24	96	5.23		
30/6	**Floweringand reproductive**	22	88	5.13	87.27^{a}	13.00^{a}
7/7		23	92	9.57		
14/7		25	100	14.75		
21/7		21	84	18.39		
28/7		19	76	21.15		
4/8		17	68	29.25		
11/8		23	92	21.10		
18/8		20	80	13.73		
25/8		24	96	5.00		
1/9		23	92	2.89		
8/9		21	84	2.04		

Means in columns followed by the same letter are not significantly different at p≤5% (Duncan's multiple range test, 1955)

Data in Table (4) showed the high content of carbohydrates in flowering and reproductive stage, while it was poor in the vegetation stage. The percent of protein contents was high through flowering and reproductive stage compared with vegetation stage. These results are in harmony with those obtained by Najafabadi *et al*. (2011), they reported that numbers of adult or immature stages of *T. urticae* showed significant variations among nitrogen treatments, where these stages of the mite increased with high nitrogen level and enhanced the mite population on bean leaves in field. These results near similar with Saleh (2017) cleared that the population of *T. urticae* was increased in the vegetation stage of soybean compared with growing stage he reported that protein content was highly in vegetation stage.

Table (4): Chemical composition of soybean crop leaves during plant growing stages.

Growing stage	Total carbohydrates%	Total potassium (K ppm)	Total phosphor P%	Total nitrogen N%	Total protein%
Vegetation	12.02^{a}	267.41^{b}	0.79^{a}	3.59^{a}	14.18^{b}
Flowering and reproductive	14.17^{a}	379.50^{a}	0.40^{b}	2.09^{b}	23.20^{a}

Means in columns followed by the same letter are not significantly different at p≤5% (Duncan's multiple range test, 1955).

Results in Table (5) conducted to determine the appropriate crop stage and the optimal number of spider mite to be initially released on soybean crop, at the age of 35 days, the bean infestation reached a maximum 102.2 individuals/leaf after 53 days followed by 92 individuals/leaf at the age of 60 days,

at the level of infection of 30 individuals/leaf. We find that at the age of 25 days for the plant we have another peak as much as 84.6 individuals/leaf after 60 days of planting, while the rest of the treatments appeared in fluctuation numbers during the growing season, which lasted up to 88 days, which started decreasing in individuals in all treatments. The treatment, 55 days was the lowest number of treatments, where, appeared 3.8 individuals/leaf at the level 10 individuals/leaf. These results near similar with Bustos *et al*. (2009) where, bean plants of four weeks of age were an excellent substrate for the development of *T. urticae* population. Generally, 35 days old soybean plants infested with spider mites at the rate of 30 mites/leaf produced significantly a greater number of spider mites. Therefore, aiming to produce a greater number of predator mites.

Table (5): Number of *Tetranychus urticae* on soybean at different crop growth stages and mite infestation densities during 2018.

Treatments		Mean number of spider mites / leaf								
Days after sowing	MD	32 Days	39 Days	46 days	53 days	60 days	67 days	74 days	81 days	88 days
25 days	10	18.40^{d}	22.60^{d}	34.20^{d}	44.20^{e}	30.60^{d}	25.40^{d}	26.80^{c}	20.60^{b}	10.20^{d}
	20	38.00^{bc}	43.40^{bc}	51.20^{c}	63.20^{c}	48.60^{c}	40.40^{c}	32.00^{b}	20.80^{b}	18.60^{c}
	30	42.60^{b}	51.40^{b}	75.00^{a}	78.20^{b}	84.60^{a}	70.00^{a}	31.20^{b}	29.60^{a}	26.20^{b}
35 days	10	26.20^{c}	29.60^{c}	40.20^{d}	52.00^{d}	36.20^{d}	28.80^{d}	30.20^{b}	24.40^{b}	19.60^{c}
	20	44.80^{b}	54.00^{b}	60.60^{b}	69.00^{b}	64.00^{b}	57.40^{b}	41.40^{a}	30.00^{a}	18.00^{c}
	30	60.00^{a}	76.20^{a}	80.80^{a}	102.20^{a}	92.00^{a}	53.40^{b}	40.00^{a}	34.80^{a}	30.60^{a}
45 days	10	14.00^{d}	20.20^{d}	26.00^{e}	34.80^{e}	20.00^{e}	18.40^{e}	14.80^{d}	16.00^{c}	09.20^{d}
	20	25.80^{c}	34.00^{c}	40.20^{d}	50.60^{d}	42.20^{c}	34.00^{c}	32.20^{b}	24.00^{b}	14.40^{c}
	30	38.00^{bc}	42.40^{bc}	52.00^{c}	56.20^{d}	62.20^{b}	63.80^{a}	24.80^{c}	34.00^{a}	36.80^{a}
55 days	10	00.00^{e}	00.00^{f}	08.20^{f}	12.80^{f}	13.40^{f}	10.00^{e}	10.60^{d}	05.20^{d}	03.80^{e}
	20	00.00^{e}	02.60^{e}	03.80^{f}	04.80^{f}	06.20^{f}	12.20^{e}	08.60^{d}	05.40^{d}	04.40^{e}
	30	00.00^{e}	00.00^{f}	00.00^{g}	07.20^{f}	06.60^{f}	11.60^{e}	10.20^{d}	11.20^{c}	09.20^{d}

Means in columns followed by the same letter are not significantly different at p≤5% (Duncan's multiple range test, 1955) MD: Soybean infested with number of spider mites / leaf.

From the predator's results in Table (6), the age of the soybean plant had no effect on the predator population. We noticed fluctuation in predator population during the growing season. However, the predator population was affected by its release rate. Three levels 2, 4 and 6 individuals were released at a fixed prey level of 30 individuals/leaf. The release rate 4 and 6 predators resulted in several individuals similar to both of levels during the growing season that extended to 88 days. The highest number of predators was 14.8 individuals at 60 days at the level of 6 predators followed by 14.4 individuals at 60 days at the level of 4 predators. The population reached 14 individuals appeared at levels 4 and 6 predators, but during different days after planting began 53 days from planting to 74 days after planting. The results obtained shows that the level of 4 and 6 predators gave the highest population during the growing season this is preferable when used and released in mite control at these levels.

Table (6): Number of *Euseius scutalis* on soybean infested with *Tetranychus urticae* at different crop growth stages during 2018.

Treatments		Mean number of spider mites / leaf								
Days after sowing	MD	32 days	39 days	46 days	53 days	60 days	67 days	74 days	81 days	88 days
25 days	2	3.40^{b}	4.60^{b}	08.00^{b}	07.00^{c}	10.00^{b}	06.60^{c}	04.40^{d}	03.20^{c}	1.20^{c}
	4	4.20^{b}	6.00^{a}	09.20^{a}	10.00^{b}	12.40^{a}	08.00^{b}	06.00^{c}	06.20^{b}	3.00^{b}
	6	5.80^{a}	6.80^{a}	10.00^{a}	11.20^{b}	14.80^{a}	10.40^{ab}	07.80^{b}	08.80^{b}	4.00^{a}
35 days	2	3.80^{b}	4.80^{b}	07.00^{b}	08.00^{c}	09.40^{b}	10.60^{ab}	06.40^{c}	03.40^{c}	2.00^{b}
	4	6.80^{a}	8.40^{a}	11.00^{a}	14.00^{a}	14.40^{a}	08.80^{b}	07.00^{b}	05.40^{b}	4.40^{a}
	6	5.40^{a}	6.80^{a}	10.00^{a}	07.60^{c}	09.00^{b}	05.80^{c}	07.40^{b}	03.80^{c}	4.60^{a}
45 days	2	2.80^{c}	5.80^{b}	11.20^{a}	12.00^{ab}	08.60^{c}	08.80^{b}	05.60^{c}	03.80^{c}	4.60^{a}
	4	4.00^{b}	5.20^{b}	09.40^{a}	12.40^{ab}	13.60^{a}	14.00^{a}	08.80^{b}	10.60^{a}	5.20^{a}
	6	5.60^{a}	8.00^{a}	09.20^{a}	09.20^{b}	11.00^{b}	12.60^{a}	14.00^{a}	07.80^{b}	4.80^{a}
55 days	2	1.40^{c}	0.00^{c}	02.60^{c}	02.80^{d}	02.20^{d}	00.00e	00.00^{f}	00.00^{d}	0.00^{d}
	4	0.00^{e}	0.00^{c}	00.00^{d}	03.00^{d}	05.00^{d}	01.60^{d}	01.80^{e}	00.00^{d}	1.00^{c}
	6	0.60^{d}	0.00^{c}	00.00^{d}	00.80^{e}	01.80^{d}	01.00^{d}	00.00^{f}	00.00^{d}	0.00^{d}

Means in columns followed by the same letter are not significantly different at p≤5% (Duncan's multiple range test, 1955) MD: Soybean infested with number of spider mites / leaf

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