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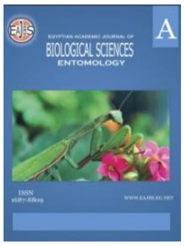

Susceptibility of Certain Egyptian Wheat Cultivars to Greenbug, *Schizaphis graminum* (Rondani) (Homoptera: Aphididae) Infestation

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ABSTRACT

The greenbug (*Schizaphis graminum*) (Homoptera: Aphididae) is a major aphid species attacking cereal crops and responsible for viral disease transmission of the plant. Host plant resistance is a strong pillar in Integrated Pest Management (IPM) for reducing the damage of this pest. This study was carried out at Plant Protection Department, Faculty of Agriculture, Al-Azhar University, Cairo, Egypt during 2019-2020 to study the susceptibility of five Egyptian wheat cultivars (Beni Suef 5, Gimmiza 11, Giza 168, Misr 1 and Sids 12) to greenbug infestation. The results indicated that in the host preference (free choose) experiment, Sids 12 cultivar was resistant (R) against aphid, while Giza 168 and Misr 1 were moderately resistant (MR) cultivars, on contrast Beni Suef 5 and Gimmiza 11 were susceptible (S) for aphid infestation. The nymphs fed on Beni Suef 5 cultivar had significantly shorter developmental time (7.6 days), highest survival (96.1%) and shorter adult longevity (9.7 days) with average offspring 45.8 aphids/ female, than other four wheat cultivars, while nymphs reared on Sids 12 had significantly longest developmental time (9.2 days), lowest survival (86.3%) and longest adult longevity (12.2 days) with average offspring 31.4 aphids/female. Feeding of *S. graminum* on tested wheat cultivars leaves significantly reduces the growth parameters (shoot length, wet and dry weights) and photosynthetic pigments content. Results from this study are important for plant breeding programs that have the objective of producing aphid resistant cultivars.

INTRODUCTION

Wheat (*Triticum aestivum*) is the most important grain worldwide is ranked second after corn based on grain acreage. In year 2019/2020 wheat was produced came to about 757 million tons in the world (FAO, 2020). Wheat is an important commodity due to its use as flour, durability and longevity. During 2020, the wheat production in Egypt amounted to approximately 8.9 million tons, with an increase of 1.48 percent from the previous year (Statista, 2020).

For over 150 years the greenbug, *Schizaphis graminum* (Rondani) (Homoptera: Aphididae), was recognized as a major pest of grains. About 40 *Schizaphis* species recognized worldwide (Blackman and Eastop, 2000). In warm or mild climates, it asexually reproduces (i.e., parthenogenesis), the female produced nymphs directly.

During seven to nine days at 16 to 27 °C, greenbugs migrate through three nymphal instars direct into the adult stage. The host range of the greenbug about 70 graminaceous species (Michels, 1986). By sucking the plant's cell sap, aphids reduce the wheat yield either directly (35-40%) or indirectly (20-80%) by transmitting fungal and viral diseases (Aslam *et al.,* 2005). Oakley *et al.* (1993) observed a reduction in wheat yield by 39%. *S. graminum* herbivory results in a significant loss in the yield of many crops.

Planting resistant cultivars to the pests infestation is a simple and effective method to decrees its damage (Shahzad *et al.,* 2019). The plant content is a key determinant of the fecundity of insect pests which affects at both the individual and the population scale **(**Awmack and Leather 2002)**.** Resistant plants may serve as a defensive mechanism against insect pests, resulting in a decrease in their reproduction and an increase in development time (Legrand & Barbosa 2000). Junaid *et al.* (2016) reported that most IPM pest control techniques have been used, i.e. biological, cultural, mechanical, physical, chemical, and the resistance of the host plant has been shown to be the best instrument in the world to decrease aphid losses. Plant tolerance to insect infestation advises the use of resistant varieties in IPM to minimise damage to insect pests. Host plant resistance can reduce the aphid population under the economic threshold level (Lowe 1987).

Therefore, the objectives of this study were to compare the susceptibility of five Egyptian wheat cultivars to the infested with aphid greenbug (*S. graminum* R.). The efficacy of integrated aphid management in the cultivation of wheat will be improved by this information.

MATERIALS AND METHODS

The present study was carried out at the Plant Protection Department, Faculty of Agriculture, Al-Azhar University, Cairo, Egypt during 2019-2020.

Wheat Cultivars:

In this study, a total of five Egyptian wheat cultivars against *S. graminum* infestation were screened. Seeds of the tested wheat cultivars; Beni Suef 5, Gimmiza 11, Giza 168, Misr 1 and Sids 12 were obtained from Agricultural Research Center, Dokki, Giza, Egypt. The wheat seeds were planted in plastic pots, filled with a mixture of soil (2:1:1 field soil, sand and rotten dung, respectively). The plants were grown in the greenhouse and then transported to a growth chamber at a temperature of 25 ±2 ºC, 60 ± 5 % RH, and exposed to a photoperiod of 14:10 L: D, light for performing the experiments. Plants were watered as necessary.

Aphids Colony:

A colony of *S. graminum* was originally obtained from the Agricultural Research Center, Dokki, Giza, Egypt. The stock colony reared in the laboratory of Plant Protection Department, Faculty of Agriculture, Al-Azhar University, Cairo. Aphids were reared on wheat seedlings of the five tested cultivars in separate (as previously described).

Experiments:

Three experiments were conducted in the Plant Protection Department greenhouse and in the growth chamber under previously described conditions to determine categories of resistance to greenbug.

1. Host Preference (Aphid free choose):

The seeds of the tested wheat cultivars were planted in a circular pattern about 3 cm from the edge into plastic pots with a diameter of 50 cm in the host selection test., this experiment was conducted during May 2020. The seedlings were cut at the same height (15 cm, in the two-leaf stage) to equalize the height to prevent an effect of plant length on the attracted aphids. Fifty individuals of *S. graminum*, were released on filter paper in the center of the pot. The seedlings were kept inside meshed cages to inhibit aphids from escaping. After 24, 48 and 72 h, counted and recorded the number of adult apterous aphids established on each cultivar. The damage done as a result of aphid infestation to each cultivar was recorded as damage rating scale of 0 to 9, where 2-3 damage rating stands for resistant (R), 4-6 damage rating represent for moderately resistant (MR) while, 7-9 damage rating for susceptible (S) varieties (Inayatullah *et al.*, 1993). Five replicates for each cultivar were used.

1.1. Development and Fecundity (No-Choose Experiment):

To evaluate the developmental time and survivorship of immature stages (nymphs) and fecundity and longevity of adults, seeds of entries were planted in 7.6 cm -diameter pots and were thinned to one seedling per pot. Individual plants in the first leaf stage were infested with one aphid adults from laboratory colonies. Each seedling covered with a plastic cage (6 cm diameter and 30 cm height) and covered with muslin cloth from the top and both sides with ventilation holes. When reproduction began, the adult was removed, and one nymph on each plant was left, which could grow on test entries until it became mature and started reproduction. Numbers of nymphs produced daily were counted and then removed from each seedling until the mature female stopped production and died (Akhtar and Mujahid, 2006). The previous parameters were estimated at ten replicates per each cultivar.

1.2. Effect of Aphid Infestation on Plant Growth and Photosynthetic Pigment Content:

The seedling of five tested wheat cultivars occurred in two groups that each cultivar had ten replicates in each group. One group received ten aphids per plant treatment and the other received no aphids. Shoot lengths of individual seedlings (in centimeters) were measured from soil to the tip of the longest leaf, just before infestation and chose the shoots to equalize on length (10 cm). Infestations were checked daily to remove or add aphids as needed to maintain 10 mature aphids/ plant and the other received group no aphids (uninfested) over 15 days period. When the symptoms of infestation appeared on plants (after 15 days), samples were taken to estimate the following measurements. Differences in shoot length, leaf area, wet & dry weights and photosynthetic pigments content between the 0 and 10 aphid plant treatments for plants of the same cultivar were measured (Razmjou *et al.* 2012). In fresh leaves chlorophyll a, chlorophyll b and carotenoid were determined by spectrophotometric method recommended by Metzner *et. al.* (1965).

Statistical Analysis:

The obtained results were subjected to analysis by SAS software; means were compared by the least significant difference (LSD) test after the significant F-test at = 0.05 (SAS Statistics, 2000).

RESULTS AND DISCUSSION

Host Preference (Aphid free choose):

Host selection or landing responses by *S. graminum*, differed among wheat cultivars shown in Table (1). The results cleared that after 24 h of aphids releasing, Sids 12 cultivar was resistant (R) to aphids infestation (1.8 aphids/ seedling after 24 h), while Giza 168 and Misr 1 were moderately resistant (MR) cultivars, on contrast Beni Suef 5 and Gimmiza 11 were susceptible (S) to aphids infestation. The same trend was recorded after 48 and 72 h. Results of our studies in agreement with those of Aly (2018) who evaluate wheat cultivars for resistance against the bird cherry-oat aphid, (*Rhopalosiphum* *padi* L.) in the laboratory. They indicated that Sids 1 was least preferred for aphids and moderately preferred cultivars were Giza 168 and Misr1, while Gimmiza 11 was highly preferred by aphids. Nazir *et al.* (2018) found that, out of 23 wheat genotypes fed against cereals aphid greenbug, five genotypes were resistant, thirteen genotypes were found moderately resistant and the rest of the five showed susceptibility to aphids. Shahzad *et al.* (2019) revealed that significant differences were recorded for *S. graminum* abundance on different wheat cultivars. Shah *et al.* (2015) and Iqbal *et al.* (2018) reported that aphid feeding differed by the plant host species and resistance mechanism shown by the plants.

Table 1: Proportion of *S. graminum* apterous adults selecting wheat cultivars after 24, 48 and 72 h per seedling (mean ±S.E.).

Wheat cultivars	After 24 h		After 48 h		After 72 h	
	Mean aphid no.	Resistance	Mean aphid no.	Resistance	Mean aphid no.	Resistance
Beni Suef 5	7.80±0.45	S	8.20±0.45	S	8.80±0.45	S
Gimmiza 11	7.20±0.84	S	7.60±0.55	S	7.00±1.22	S
Giza 168	4.40±0.55	MR	5.00±0.71	MR	5.40±0.55	MR
Misr 1	5.00±1.00	MR	5.40±0.89	MR	5.80±0.45	MR
Sids 12	1.80±1.80	R	2.60±0.55	R	2.80±0.45	R
$LSD_{5\%}$	0.91*		0.85*		0.91*	
F value	59.87		59.62		50.70	
P value	0.000		0.000		0.000	

The values are presented as means ± SE of five replicates. * significant differences among treatments ($P < 0.05$, ANOVA). R (resistant), 2-3; MR (moderately resistant), 4-6; S (susceptible), 7-9 (Inayatullah *et al.*, 1993).

In the very first step of host selection, the decision on the suitability of the plant as a host is made. Aphids use both visual and chemical signals (Döring and Chittka, 2007) (Pickett et al., 2007) to decide to land on a plant. Trichomes are the first line of defence when aphids land. Trichomes may either be non-glandular or glandular. Trichome density has a major effect on aphid movement and feeding, independent of their composition (Musetti and Neal, 1997 and Bin, 1982). Plants also have other constitutive defences that offer direct resistance to plants against aphid feeding, such as thick cell walls. Although these mechanical barriers are constitutive defences, they can also be created (directly induced defences) in response to aphid feeding (Kumar, 2019). Mithöfer and Boland (2012) proposed that plants have developed a plethora of various chemical defences for herbivores of all kinds that are poisonous, repellent or antinutritive.

Development and Fecundity (No-Choose):

1.Developmental Time and Survivorship of Nymphs:

Table (2) shows the results of the developmental time of *S. graminum* nymphal stage, significant differences among the five tested Egyptian wheat cultivars. The nymphs reared on Beni Suef 5 cultivar (S) had significantly shorter development time (7.6 days) than the other four wheat cultivars. While nymphs reared on Sids 12 (R) had significantly the longest development time (9.2 days). The percentage of nymphal survival varied from highest on Beni Suef 5 (96.1%) to lowest on Sids 12 (86.3%). These results in agreement with Taheri *et al.* (2010) and Aly (2018) who found that the developmental time and mortality of nymphal stage of aphids indicated significant differences among the examined wheat varieties.

Table 2: Nymph developmental time, nymph survival, adult longevity, and total no. of offspring/ female of *S. graminum* on five tested Egyptian wheat cultivars.

Wheat cultivars	Nymph developmental time (day)	Nymph survival (%)	Adult longevity (day)	Total no. of offspring/ female
Beni Suef 5	7.60±0.70	96.1	9.70±0.67	45.80±2.20
Gimmiza 11	8.40±0.70	95.8	10.60±1.26	44.10±0.99
Giza 168	8.50±0.71	93.4	10.90±1.20	45.10±1.73
Misr 1	8.80±0.63	93.3	11.10±0.99	42.50±2.12
Sids 12	9.20±0.79	86.3	12.20±1.03	31.40±2.63
$LSD_{5\%}$	0.64 *		0.94*	1.81*
F value	7.00		7.34	86.91
P value	0.000		0.000	0.000

The values are presented as means ± SE of ten replicates. * significant differences among treatments ($P < 0.05$, ANOVA).

2.Adult Longevity and Reproductive Ability:

Adult longevity was the shorter in aphids reared on Beni Suef 5 cultivar (9.7 days) in comparison to aphids reared on other cultivars. Aphids that fed on Sids 12 showed the longest longevity (12.2 days). Mean fecundity recorded in *S. graminum* adults fed on different wheat cultivars was found to be significantly different. Aphids fed on leaves of Beni Suef 5 cultivar produced average offspring of 45.8 aphids/ female, however 44.1, 45.1, 42.5 and 31.4 aphids/female for aphids reared in Gimmiza 11, Giza 168, Misr 1 and Sids 12 respectively with a significant difference between cultivars. The acquired data cleared that different wheat cultivars examined in the present work differed as hosts for *S. graminum* in terms of their quality, an insect pest of wheat. The results are consistent with previous findings by Castro *et al.* (1999), Hu *et al.* (2016) and Hu *et al.* (2018) who found that wheat varieties affected the biological parameters i.e. developmental time and longevity and fecundity of the aphids. The biology of aphids on various cultivars needs to be well researched in order to provide the necessary knowledge to develop aphid management strategies in a specific area (Xia *et al.* 1999 and Razmjou *et al.* 2006).

Effect of Aphid Infestation on Plant Growth and Photosynthetic Pigments Content:

1. Growth Parameters:

The growth parameters; shoot length (cm), leaf area (cm^2), wet and dry weights (g/plant) of wheat cultivars were affected by ten aphids per plant infestation with *S. graminum* during a 21days period. Differences in growth parameters of each cultivar were used as a dependent test for tolerance. However, aphid infestation level reduced all these parameters in infested plants with respect to uninfested plants. But their differences in the percentage of reduction in shoot length, wet and dry weights were significantly affected, while differences in leaf area were no significant. Sids 12 cultivar was significantly tolerant than the other four cultivars (Table 3). The results are in agreement with Razmjou *et al.* (2012) and Mojahed *et al.* (2013) who found that the growth parameters; shoot length, root length, wet and dry weights of wheat lines were affected by the infestation of aphids. The health of the host plant is typically determined by aphid feeding, and it is suspected that greenbug has substances that are harmful to the host plant in its saliva, affecting the growth of root and shoot mass and tillering in wheat (Burton and Burd 1993; Riedell and Kieckhefer 1995). Feeding of aphids contributes to the removal of significant amounts of plant sap, resulting in local chlorosis, plant weakness (Kumar, 2017). In countering plant defence response and changing the incompatible relationship to a compatible one by modifying the metabolism of plants, aphid saliva plays an important role. Aphid feeding can lead to changes in host plants, including changes in morphology, alteration of resource allocation, and local and systemic symptom development (Walling, 2008).

Table 3: Percent change of wheat cultivars in growth parameters with *S. graminum* infestation.

Wheat cultivars	Shoot length (%)	Leaf area (%)	Wet weight (%)	Dry weight (%)
Beni Suef 5	21.78	12.45	18.51	14.53
Gimmiza 11	17.98	11.58	16.37	13.41
Giza 168	15.97	10.47	13.66	11.41
Misr 1	12.56	9.45	12.04	11.81
Sids 12	11.59	7.85	11.67	9.10
LSD $_{5\%}$	6.12 *	5.18ns	3.59*	0.016*
F value	3.98	1.05	5.84	11.80
P value	0.015	0.211	0.002	0.000

The values are presented as a percent change of ten replicates. (*) significant, (ns) no differences among treatments (P < 0.05, ANOVA).

2. Changes in Photosynthetic Pigments Content:

As shown in Table (4) feeding of *S. graminum* on all wheat cultivars significantly reduced the chlorophyll a, b and carotenoid content. Feeding on Beni Suef 5, Gimmiza 11, Giza 168, Misr 1 and Sids 12 led to reduced chlorophyll (a) contents by 23.38, 22.01, 19.19, 20.60 and 17.03% respectively, as compared to the corresponding control. In the same order, chlorophyll (b) contents were reduced by 20.69, 19.06, 15.52, 17.04 and 14.39% respectively, with significant differences (P=0.021 and 0.008) between tested cultivars in chlorophyll content. While, the carotenoids contents were depressed by 16.16, 15.12, 15.17, 13.70 and 12.19% in the same respect, with non-significantly differences (P=0.151) between tested cultivars. The results are in harmony with those obtained by Shahzad *et al.* (2019); Zhang *et al.* (2019); Liu *et al.* (2020) and Zhang *et al.* (2020) who found that feeding of *S. graminum* on wheat leaves significantly reduce the chlorophyll content. Al-Mousawi *et al.* (1983) Aphid feeding has been proposed to change physiological and biochemical processes, such photosynthesis, in plants. For example, feeding *S. graminum* has caused damage to cell walls and chloroplasts due to saliva degrading enzymes. Forrest (1971); Sandström *et al.* (2000) and Cao *et al.* (2016) showed that aphids in their host plants are able to regulate nutritional supply and quality. The enzymes in aphid saliva can break down leaf chloroplasts, leading to white, yellow, purple, or red-purple longitudinal streaks on leaves of the infested plants (Ma *et al.* 1998).

Table 4: Percent change of chlorophyll a & b and carotenoid contents (mg/g fresh wt.) with *S. graminum* infestation for five Egyptian wheat cultivars.

Wheat cultivars	**Chlorophyll a (%)**	**Chlorophyll b (%)**	**Carotenoids (%)**
Beni Suef 5	23.38	20.69	16.16
Gimmiza 11	22.01	19.06	15.12
Giza 168	19.19	15.52	15.17
Misr 1	20.60	17.04	13.70
Sids 12	17.03	14.39	12.19
LSD $_{5\%}$	3.81	3.51	3.32
F value	3.64	4.63	1.89
P value	0.021 *	0.008 *	0.151 ns

The values are presented as a percent change of ten replicates. (*) significant, (ns) no differences among treatments (P < 0.05, ANOVA).

Fouché *et al.* (1984); Burd and Burton (1992) who reported that chlorophyll deficiency due to aphid infestation in susceptible varieties reduces yields by up to 50%. Burd and Elliott (1996) and Rafi *et al.* (1996) mentioned that Russian wheat aphid, feeding results in destruction of plant chloroplasts that ultimately lead to reduced chlorophyll levels and photosynthetic activity. This decrease in chlorophyll showed that aphid feeding had a detrimental effect on the plant and had a direct impact on the chlorophyll amount (Heng-Moss *et al.*, 2003). Ciepiela, (1993) showed that aphids inject into the plants harmful saliva that eventually prevents the formation of grain.

Conclusion

Out of five wheat cultivars tested, Sids 12 cultivar was more tolerant against the aphid infestation. The use of this cultivar can be an economical method to managing *S. graminum* damage in wheat crops and reduce the yield losses. Also, this tolerant cultivar may help to minimize the possible use of insecticides and to improve future integrated pest management programs. It is recommended to further evaluate this cultivar for its tolerance against aphids under field conditions as a potential candidate for less aphid infestation.

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ARABIC SUMMARY

قابلية بعض أصناف القمح المصري للإصابة بمنّ القمح *Schizaphis graminum*

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يعد منّ القمح من الافات الخطيرة التى تهاجم محاصيل الحبوب النجيلية ومسؤول عن إنتقال الأمراض الفيروسية والفطرية للنبات. وتعتبر مقاومة النبات للإصابة الحشرية ركيزة أساسية في الإدارة المتكاملة للآفات (IPM) للحد من أضرارها. لذا أجريت هذه الدراسة في قسم وقاية النبات بكلية الزراعة جامعة الأزهر بالقاهرة خلال موسم 2019-2020 لدراسة مدى قابلية خمسة أصناف من القمح المصري (بني سويف 5 ، جميزة 11 ، جيزة 168 ، مصر 1 وسدس 12) للإصابة بمنّ القمح.

أشارت النتائج في تجربة تفضيل العائل (الاختيار الحر) ، أن الصنف سدس 12 أكثر الأصناف مقاومة ضد الإصابة بالمنّ، بينما كان الصنف جيزة 168 و مصر 1 متوسطي المقاومة، على النقيض كان الصنف بني سويف 5 و جميزة 11 أكثر قابلية للإصابة بالمن. كانت مدة التطور للحوريات التي تغذت على صنف بني سويف 5 أقصر (7.6 يوم) وأعلى معدل بقاء (96.1 ٪) وأقصر مدة للطور اليافع (9.7 يوم) بمتوسط نسل 45.8 حورية لكل حشرة منّ، مقارنة بلاربعة أصناف القمح الأخرى، بينما الحوريات التى ربيت على الصنف سدس 12 أخذت واقت أطول حتى بلوغ الطور اليافع (9.2 يوم)، وأقل نسبة بقاء (86.3٪) وأطول عمر للبالغين (12.2 يومًا) بمتوسط ذرية 31.4 حورية لكل حشرة منّ. إن تغذية المنّ على أوراق القمح المختبرة قلل بشكل معنوى من طول الساق، الوزن الرطب والجاف ومحتوى النبات من الصبغات. تعتبر نتائج هذه الدراسة مهمة لبرامج تربية النباتات التي تهدف إلى إنتاج أصناف مقاومة لحشرات المن.