# Resistance of Wheat lines to Greenbug, *Schizaphis graminum* (Rondani) (Hemiptera: Aphididae)

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# ABSTRACT

The greenbug *Schizaphis graminum* (Rondani) is one of the most important cereal pests in the world. Therefore, we conducted two sets of experiments for evaluating of resistance of wheat lines against this aphid. An initial screening test assessed resistance in forty wheat lines to this aphid in laboratory conditions, and then biological characteristics of this pest were evaluated on eight of these. Test aphids were reared on Hirmand wheat at  $25 \pm 1^{\circ}$ C,  $60 \pm 5$  % R.H. and a photoperiod of 14: 10 h (L: D). The developmental time of immature stage ranged from 6.0 to 7.0 day. Total offspring per female ranged from 33.9 to 60.4 nymphs on N88-19 and N88-8 lines, respectively. Offspring produced per female per reproduction day was the highest on N88-11 (3.3 nymphs) and lowest on N87-8 and N87-9 (2.8 nymphs). The intrinsic rate of natural increase ( $r_m$ ) varied from 0.251 on N88-19 to 0.321 on N88-11. The  $R_o$  values ranged from 16.3 on N88-19 to 52.4 on N88-8. The other life table parameters such as  $\lambda$  (finite rate of increase), *T* (mean generation time) and *DT* (doubling time) were also calculated. The results of this study indicated that N88-19, N87-11, N87-8 and N87-9 wheat lines are partially resistant against this pest and could be used in integrated pest management (IPM) programs.

Key words: Biological characteristics, green aphid, plants resistance, wheat lines.

# INTRODUCTION

The greenbug, *Schizaphis graminum* (Rondani), is an important pest of Graminae crops, especially small grains with worldwide distribution (Al-Mousawi *et al.*, 1983; Blackman and Eastop, 2000; Modarres Najafabadi, and Gholamian, 2006; Tofangsazi *et al.*, 2012). Its host range includes a wide range of cultivated and noncultivated grass species (Michels, 1986) so that important cereals such as wheat, barley, maize, and sorghum are attacked severely by this aphid (Weng *et al.*, 2010). *S. graminum* imposes direct and indirect damages to cereals by feeding on plants and virus transmission. The aphids inject salivary secretions into plants which break down chloroplasts and cell walls in susceptible plants (Al-Mousawi *et al.*, 1983), causing chlorosis and reduced photosynthetic rates. Many plant viruses including barley yellow dwarf virus (Murphy, 1959), sugarcane mosaic virus (Ingram and Summers, 1938), and maize dwarf mosaic virus (Nault and Bradley, 1969) are transmitted by greenbug, increasing the problems caused by this aphid. Consequently, combination of aphid feeding on

plants and transmission of plant viruses lead to enhance crop damages. Severe damage from greenbug feeding (Rezvani, 2001) necessitates surveying resistant levels of wheat plant to this aphid. Additionally, pesticide resistance problems support the use of resistant or tolerant lines or varieties against S. graminum. On the other hand, understanding effects of host plant on development and reproductive characteristics of phytophagous insects provides insight into the evolution of phyto-phytophage relations, and helps in predicting population outbreak as well as improves pest management approaches. Development, fecundity and survival of phytophagous insects are influenced by host plant quality (Awmack and Leather, 2002). Suitability of different cultivars or lines of the host plant for the aphids and its effect on their population growth can be investigated by estimating biological parameters, particularly the intrinsic rate of increase  $(r_m)$ . Several scientists have reported the reproductive characteristics of different greenbug biotypes on various host plants such as sorghum (Gorena, 2004), wheat varieties (Fattah Alhoseini et al., 2010), and barley (Tofangsazi et al., 2012). In addition, resistance or susceptibility of wheat varieties to another wheat aphid, Sitobion avenae were evaluated by Razmjou et al., (2011).

Therefore, due to the importance of *S. graminum* in cereal production, as a serious threat to these plants, more studies have to be performed on the effect of different agents on development and reproductive aspects of this pest in Iran and elsewhere. Hence, the objective of this research was to assess the effect of eight wheat lines, identified by an initial screen of 40 wheat lines, on development and reproductive characteristics of *S. graminum*. These findings would help to promote the level of managing of wheat production.

# MATERIALS AND METHODS

#### **Plant materials**

Among all of the wheat (*Triticum aestivum*) lines planted in the research station of the Agriculture and Natural Resources Center of Moghan, 40 lines [N87 (1-20) and N88 (1-20)] were selected for initial screening test against *S. graminum*. Seeds were obtained from Agriculture and Natural Resources Research Center of Moghan, Iran. Disinfected seeds (five seeds per pot) of these lines were planted in 7-cm plastic pots. All of the tested plants were reared in a greenhouse placed at the Faculty of Agriculture, University of Mohaghegh Ardabili, Iran.

## Aphid colony

Greenbugs obtained from Faculty of Agriculture, Tarbiat Modares University were reared on seedling of Hirmand wheat variety in a growth chamber at  $25 \pm 1$  °C,  $60 \pm 5\%$  R.H. and a photoperiod of 14: 10 h (L: D) in December 2010. Aphids were reared for several generations (1 month) before initiating of experiments.

#### **Initial Screening test**

Two-to-three-leaf stage plants were placed in a growth chamber at a temperature of  $25 \pm 5$  °C,  $65 \pm 10\%$  R.H. and a photoperiod of 14: 10 h (L: D). Each wheat line was planted in 4 replicates. Five seeds of each line were sown in a 7-cm plastic pot. At the two or three-leaf stage of development, pots were thinned to one plant and then two adult aphids were transferred to each plant. After 24 hours, all aphids except three nymphs were removed. These nymphs were allowed to feed on the seedlings for 14 days. The aphids on each seedling were then counted. Two wheat lines died prematurely so only 38 lines of wheat were finally evaluated.

#### Life table experiments

Eight wheat lines were selected on the basis of screening test for further assays of aphid performance. These lines were selected as described above. Lines included N88-3, N88-8, N87-8, N87-9, N87-11, N88-11, N88-14 and N88-19. Two randomly selected apterous aphids from stock culture were placed on the leaf surface (at the two or three-leaf stage of development) inside cylindrical plastic cages (6 cm in diameter, 30 cm in height) to prevent escape or parasitism. After 24 h all but three nymphs were removed from each plant and remaining nymphs were monitored daily to assay development and fecundity rates. After maturity, nymphs produced per reproduction day were recorded daily. Newborn nymphs were removed. There were 21-25 replicates (plants) for each line.

#### Statistical analysis

Data on the number of aphids in screening test were evaluated using ANOVA. When the mean values indicated significant differences among treatments, they were separated by the tukey's test at  $\alpha$  = 0.05 (SAS Institute, 2000). Mean number of S. graminum produced per wheat line are shown in Fig. 1. We evaluated developmental time and survival of the nymphal stage, adult longevity and fecundity. Data on these parameters after normality test by Minitab ver. 15 statistical software (Minitab Inc, 1994), were analyzed by one-way ANOVA using SPSS ver. 16.0 (SPSS, Chicago, IL). Total fecundity, reproduction period and adult longevity data were normal scores transformed to stabilize the variance before analysis. When the mean values indicated significant differences among treatments; they were separated using tukey's test at probability level of 0.05 (SAS Institute, 2000). Also, life table parameters, including the intrinsic rate of natural increase  $(r_m)$ , net reproductive rate  $(R_o)$ , doubling time (DT), finite rate of increase ( $\lambda$ ) and mean generation time (T), were calculated by the jackknife method (Meyer et al., 1986; Carey, 1993) using Maia's program written for the SAS system (SAS Institute, 2000; Maia et al., 2000). The pseudo-values of life table parameters at first were tested for normality using Minitab 15 software (Minitab Inc., 1994) and then were analyzed by one-way ANOVA using SPSS ver. 16.0 (SPSS, Chicago, IL). The data on the R<sub>o</sub> and DT for each line were normal scores transformed and T was log 10 transformed to normalize the variance before data analysis.



Fig. 1. Mean of aphid, Schizaphis graminum number on 38 wheat lines.

# RESULTS

#### Laboratory screening

Fourteen days after infestation, the number of aphids per plant differed among 38 wheat lines screened (F = 1.67; df = 37, 113; P < 0.021). The average number of aphids after 14 days is shown in Fig.1. On the N88-19, N87-11, N87-8 and N87-9 were fewer aphids that they are therefore partially resistant wheat lines against *S. graminum*.

#### Developmental time and survival of the nymphs

The mean number of days from birth to first reproduction differed (F = 9.6; df = 7, 175; P < 0.01) among the wheat lines tested. Aphids on N87-9, N88-3, N88-8, N87-8 and N87-9 were generally found to have the longest developmental time, while the shortest times were on N88-11 and N88-14 (Table 1). The percentage of nymphal survival ranged from % 48 on N88-19 to % 92 on N88-14 (Table 1).

Wheat lines		Develop	ment data (mean ±	Fecundity data (mean ± SE)		
	N	Nymphal survival (%)	Developmental time (days)	Adult longevity (days)	Total no. of offspring/female	No. of offspring/ reproduction day
N88-3	23	79	6.8 ± 0.08ab	30.9 ± 2.1a	58.3 ± 3.2a	3.0 ± 0.1a
N88-8	21	87	6.8 ± 0.08ab	29.3 ± 2.1a	60.4 ± 4.0a	3.2 ± 0.1a
N87-8	22	61	6.9 ± 0.1ab	15.1 ± 2.2bc	34.7 ± 4.8b	2.8 ± 0.2a
N87-9	22	67	7.0 ± 0.0a	24.0 ± 2.4ab	45.3 ± 4.1ab	2.8 ± 0.1a
N87-11	22	56	6.8 ± 0.08ab	23.0 ± 2.2abc	51.2 ± 4.2ab	3.0 ± 0.1a
N88-11	25	76	6.0 ± 0.0c	25.4 ± 2.1ab	55.8 ± 3.7a	3.3 ± 0.1a
N88-14	24	92	6.4± 0.1bc	21.9 ± 2.7abc	45.9 ± 4.8ab	3.1 ± 0.1a
N88-19	24	48	6.6 ± 0.22ab	15.1 ± 2.5c	33.9 ± 5.2b	2.9 ± 0.2a

Table 1. Means (± SE) of development data and fecundity of S. graminum on eight wheat lines.

For each parameter, differences among wheat lines were determined by Tukey's tests. Within columns, means followed by different letters are significantly different (P < 0.05).

#### Longevity and fecundity

Significant effects were observed for both longevity (F = 6.77; df = 7, 175; P < 0.01) and total number of offspring per female (F = 5.26; df = 7, 175; P < 0.01). Means for the total number of offspring per female were highest on N88-8, N88-3, and N88-11 and lowest on N87-8 and N88-19 lines, respectively (Table 1). However, it was highest on N88-8 and lowest on N88-19. The number of offspring produced per female per reproduction day by *S. graminum* indicated no significant differences (F = 1.56; df =7, 175; P < 0.15) among wheat lines (Table 1; Fig. 2). Nymphal survival was highest on N88-14 and lowest on N88-19. The survival rate of nymphs developing to adults from the initial cohort was estimated 48, 56, 61, 67, 76, 79, 87 and 92 % on N88-19, N87-11, N87-8, N87-9, N88-11, N88-3, N88-8 and N88-14, respectively. Death of the last female (maximum age) occurred at the ages of 41, 45, 42, 44, 42, 45, 45 and 44 days, respectively (Fig. 2).



Fig. 2. Survival rate ( $I_x$ ) and fecundity ( $m_x$ ) of Schizaphis graminum on eight wheat lines at 25°C.

#### Life table parameters

The intrinsic rate of natural increase ( $r_m$ ) values of the apterous aphids differed (F = 14.14; df = 7, 175; P < 0.01) among the wheat lines examined. The  $r_m$  values

ranged from 0.251 to 0.321 nymphs/female/day (Table 2). *S. graminum* reared on N88-19 and N87-11 had the lowest  $r_m$ , and those reared on N88-11, N88-14 and N88-8 the highest. The  $R_o$  values also differed among wheat lines (F = 17.18; df = 7, 175; P < 0.01). Aphids on N88-8 and N88-3 had the largest  $R_o$  values and those on N88-19 had the lowest (Table 2). *T* values also differed among wheat lines (F = 3.55; df = 7, 175; P < 0.01) (Table 2). *DT* values of *S. graminum* also differed (F = 10.31; df = 7, 175; P < 0.01). DT values were highest on N88-19 and N87-11 and lowest on N88-11 and other lines (Table 2). The  $\lambda$  values were lowest (F = 14.30; df = 7, 175; P < 0.01) on N88-19, N87-11 and highest on N88-11, N88-14 and N88-8 (Table 2).

Parameters (mean ± SE) <sup>a, b</sup>										
Wheat Lines	N⁵	Net reproductive rate (R <sub>0</sub> )	Intrinsic rate of increase (r <sub>m</sub> )	Mean generation time ( <i>T</i> )	Doubling time ( <i>DT</i> )	Finite rate of increase ( $\lambda$ )				
N88-3	23	46.0 ± 2.5a	0.304 ± 0.005ab	12.6 ± 0.2a	2.3 ± 0.03bc	1.355 ± 0.006ab				
N88-8	21	52.4± 3.6a	0.318 ± 0.007a	12.5 ± 0.3a	2.2 ± 0.05c	1.374 ± 0.01a				
N87-8	22	21.2 ± 2.9de	0.273 ± 0.008bc	11.2 ± 0.4ab	2.5 ± 0.08ab	1.313 ± 0.011bc				
N87-9	22	30.4 ± 2.8bcd	0.273 ± 0.005bc	12.5 ± .03a	2.5 ± 0.04ab	1.313 ± 0.006bc				
N87-11	22	28.8 ±2.4cd	0.265 ± 0.005c	12.7 ± 0.4a	2.6 ± 0.05a	1.303 ± 0.007c				
N88-11	25	42.5 ± 2.8abc	0 321 ± 0.007a	11.7 ± 0.3ab	2.2 ± 0.04c	1.378 ± 0.009a				
N88-14	24	41.8 ± 4.4ab	0.315 ± 0.007a	11.9 ± 0.4ab	2.2 ± 0.05c	1.370 ± 0.01a				
N88-19	24	16.3 ± 2.5e	0.251 ± 0.012c	11.1 ± 0.7b	2.8 ± 0.13a	1.286 ± 0.015c				

Table 2. Life table attributes of S. graminum reared on eight wheat lines.

Within columns means followed by different letters are significantly different (P < 0.05).

a. For each parameter, differences between wheat lines were determined by Tukey's test.

b. N the sample size of each parameter value.

### DISCUSSION

In the initial screening, tested wheat lines differed with respect to the average aphids per plant (Fig. 1). Razmjou et al., (2011) carried out screening test of 23 wheat varieties to English grain aphid, Sitobion avenae with similar results. In the present study, fecundity rate differed among the eight wheat lines whereas, Fattah Alhoseini et al., (2010) found similar life history data, except for developmental time, for S. graminum on five wheat cultivars. This difference likely results from different thermal conditions or diverse varieties. In their study, aphid developmental time ranged from 6.36 to 7.39 days, similar to our findings. Aphids fed on N87-9 and some other lines had comparatively longer duration of nymphal stage than the other lines (7 day; Table 1). Longer duration of the nymphal stage on N87-9 and other related lines (Table 1) leads to a longer lifespan, usually lower reproductive ability and slower population increase. Among the tested wheat lines adult longevity of S. graminum were significantly different, it was longer on N88-3 and N88-8 in comparison with the other lines tested, demonstrating a higher reproductive potential for the aphids on this two lines. Our surveys indicated that total fecundity (number of offspring per female) of S. graminum was the highest on N88-8, N88-3 and N88-11, indicating favorability

of this line than the others. The intrinsic rate of natural increase  $(r_m)$  is suitable for comparing the effects of plant varieties or lines on aphid biological characteristics. Highest r<sub>m</sub> values of S. graminum were on N88-8, N88-11 and N88-14 and the lowest content refereed to the cohorts that fed on N88-19 and N87-11. The suitable r value of viviparous aptera found on N88-11 compared to the other wheat lines, likely was due to the greater fecundity and longer adult longevity of this aphid fed on this line. These values are close to those estimated for the S. graminum reared on five wheat varieties ( $r_m$  values ranged from 0.276 to 0.312 day<sup>-1</sup>) (Fattah Alhoseini *et al.*, 2010). In addition, the lower r<sub>m</sub> value of S. graminum on N88-19 was because of the poor fecundity, partially longer developmental time and higher rate of nymphal mortality (Tables 1 and 2). Gorena (2004) concluded that the  $r_m$  values of different biotypes of S. graminum on sorghum ranged from 0.16 to 0.38 and our values (0.251-0.321 day<sup>-1</sup>) fall within this range. The reasons of the wider range they observed may be due to different test conditions or, more likely, host differences. Thus, S. graminum performance was significantly better on N88-11, N88-8 and N88-14 than the other lines. Whereas, aphid poor performance was on N88-19 and N87-11, these therefore were relatively resistant to S. graminum. We did not investigate the functional basis of this occurrence, but Thackray et al., (1990) indicated that some phytochemicals, such as DIMBOA, affect wheat resistance to aphids through antibiosis and feeding avoidance. Hydroxamic acids are natural defensive agents against insects (Escobar and Niemeyer, 1993). The most abundant hydroxamic acid in wheat plants is DIMBOA (Niemeyer and Perez, 1995). The level of this substance in plants is related to insect resistance and allelopathic effects of cereals (Copaja et al., 1991). Further research is needed to determine if these compounds are involved in the resistance differences observed in our study for greenbug development, survivorship and reproduction.

Indeed, among the available control methods, host plant resistance is an important foundation of integrated pest management (IPM) (Zehnder *et al.*, 2007). Even partially resistant cultivars may improve the efficiency of biological and chemical control methods (van Steenis and El-Kawass, 1995; Du *et al.*, 2004). Hence, our findings may provide useful information to IPM programs for *S. graminum* in wheat production.

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