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Population dynamics and biological parameters of *Aphis fabae* Scopoli on five broad bean cultivars

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Abstract

Understanding how host-plant characteristics affect biological and demographic parameters of insect herbivores is of considerable importance in the development of resistant crop cultivar. Biological and life table parameters of the black bean aphid, *Aphis fabae* Scopoli (Homoptera: Aphididae), were estimated on five broad bean, *Vicia faba* L. (Fabales: Fabaceae) cultivars (4, 11, 12, 13 and 14) at 17 ± 5°C, 70 ± 10% RH and a photoperiod of 14: 10 (L: D)h. The experiment was arranged in the randomized complete block design (RCBD) and there were eight replications (plants) for each treatment (cultivar). The nymphal rate mortality of *A. fabae* was significantly different among cultivars ($P = 0.028$; $\chi^2_{9.48} = 10.91$). The cultivar 12 induced the highest mortality rate (26.83%), whereas the cultivar 4 caused the lowest mortality rate (4.65%). The results showed significant differences among the five cultivars for the following biological parameters: the adult size ($F_{3.36} = 3.391$; $P = 0.049$), the adult weight ($F_{3.18} = 3.260$; $P = 0.047$) and the total number of embryos ($F_{3.48} = 5.513$; $P = 0.013$). The poor performances of *A. fabae* were recorded on the cultivar 12 (2.54 mm^2 of size, 0.87 mg of weight and only 20.33 embryos in mean) in comparison to other cultivars. The number of developed embryos and all demographic parameters of black bean aphid were not significantly different between the cultivars ($P > 0.05$). The partial correlation showed no relationship between all biological parameters tested except the total number of embryos and the number of developed embryos; there was a positive low correlation between the two parameters ($r = 0.568$; $P = 0.03$). The present study revealed that the resistant cultivar 12 have insecticidal power and a negative effect on the performances of *A. fabae* adults and could be used as an alternative to conventional synthetic insecticides in an integrated pest management program to *A. fabae*.

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Introduction

Broad bean, *Vicia faba* L. (Fabales: Fabaceae), is one of the oldest cultivated field crops in the world. It constitutes a major protein source for human population in many countries. It is the most important food legume crop in Algeria with 4 138 900 tons of dry matter, where it covers about 1 103 700 ha (FAO stat, 2014). In Algeria, broad bean production includes highly diversified local cultivars. About 68 locally cultivars, have been identified by morphological and agronomic characterization (Meradsi, 2009).

The black bean aphid, *Aphis fabae* Scopoli (Homoptera: Aphididae), is the most destructive pest of broad bean around the world. It is one of the 14 aphid species of most agricultural importance. *A. fabae* is a very polyphagous species, but the actual host range of the aphid that colonizes beans and sugar beet is unclear, because it is a number of a bewildering complex of species, at least some of which also have wide host ranges (Blackman and Eastop, 2007). *A. fabae* occurs in Europe, Western Asia, Africa, and South America. It is a vector of more than 30 plant viruses, including non-persistent viruses of bean and peas, beets, crucifers, cucurbits, dahlia, potato, tomato, and tulip, and the persistent Beet Yellow Net Virus (BYNV) and Potato Leaf Roll Virus (PLRV) (Blackman and Eastop, 2007).

Insects are usually controlled by insecticides. However, the results of the uses of insecticides to control these pests among others, the development of high levels of resistance to conventional insecticides (Ogendo *et al.,* 2003), the high costs of synthetic pesticides and associated toxicity risks (Mihale *et al.,* 2009), the destruction of beneficial insects (pollinators, parasitoids and predators), pesticide residue magnification in humans and wildlife and disruption of ecosystem (Ruchika and Kumar, 2012), have increased the need to search for alternative insect control methods. Host plant resistance has been used as a control measure for various agricultural pests for many years (Smith, 1989 & 2005). Recently, some progress has been archived in screening broad bean cultivars against black bean aphid and cowpea aphid, and a number of cultivars have been identified as possible source of resistance for breeding programs (Khelfa, 2004; Meradsi, 2009; Lebbal, 2010; Meradsi and Laamari, 2016).

The aim of this study was to evaluate the level of resistance of five locally broad bean cultivars against the most pest of bean witch as the black bean aphid, *A. fabae*.

The biological and demographic parameters of this aphid were used to determine the most resistant cultivar in greenhouse conditions. The work was aimed to search a source of sustainable alternatives to synthetic insecticides for controlling the black bean aphid without affecting the environment, beneficial organisms or men.

Materials and methods

Host plant material

Broad bean (*Vicia faba* L.) cultivars used in this study were selected on the basis of field evaluation for resistance to *A. fabae* (Meradsi and Laamari, 2016). They included five cultivars: four resistant cultivars, 1, 12, 13, and 14; and one susceptible cultivar 4. The seeds of the cultivars were individually grown in plastic pots (13 cm diameter × 14 cm) under greenhouse conditions of 17 ± 5°C, 70 ± 10% RH and 14: 10 (L: D) h photoperiod.

Source of aphids

The aphids used in this study were obtained from a single winged adult of *Aphis fabae*. The latter was collected in early February, 2014 from broad bean plants in a field situated at the region of Biskra (in the east of Algeria). The aphids were reared on broad bean seedlings (*V. faba*) under greenhouse conditions of 17 ± 5°C, 70 ± 10% RH and 14: 10 (L: D) h photoperiod.

Biological parameters of Aphis fabae

The experiment was conducted under greenhouse conditions of 17 ± 5°C, 70 ± 10% RH and 14: 10 (L: D) h photoperiod. Antibiotic resistance was assessed using seedlings grown individually in plastic pots (13 cm diameter × 14 cm height). The test was arranged in the randomized complete block design (RCBD) and there were eight replications (plants) for each treatment (cultivar). One apterous adult was placed on each plant at 12 growth stage (two leaves unfolded) (Mier, 2001), and was allowed to deposit nymphs (between 5 and 8 larvae). After 24h, we removed the adults.

Once the larvae have reached the adult stage, an individual maintained on each plant, the others are removed, the nymphal mortality rate was determined. Once the others larvae reached the adult stage, their size (mm^2) was calculated using the formula proposed by Taylor (1975); [the 'size' given is the product of length (from frons to base of the cauda) × width (at the widest point of the abdomen)]. On the day of adult moult, females were individually weighed on a Sartorius *Basic* balance with a sensitivity of 0.1mg.

Dissection of aphids

These same adults who have not started giving larvae were also used to determine their potential fecundity. In a drop of methylene blue and under a dissecting microscope, he proceeded to dissecting each female. The ovarioles were obtained by pulling the terminal abdominal segment with a fine needle, whilst holding the head and the thorax with another needle. The total number of embryos, as well as the number of embryos with red pigmented eyes (mature embryos) was counted. The classification of embryos is based on size. The embryos with red eye spots were considered to be developed (Leather and Welling, 1981; Llewellyn and Brown, 1985; Sauge *et al.,* 1998; Traicevski and Ward, 2002).

Relationship between biological parameters

A partial correlation model was adopted to determine the nature of the relationship between each pair *viz.* Adult weight - adult size; Adult weight - total number of embryos; Adult weight - number of developed embryos; Adult size - total number of embryos; Adult size - number of developed embryos; Total number of embryos - number of developed embryos.

Demographic parameters of Aphis fabae

The intrinsic rate of natural increase (r_m) for apterous aphids on different cultivars was estimated using the following equation: $r_m = 0.74\ (\ln Md)/d$ (Wyatt and White, 1977), where d is the developmental time from birth to onset of reproduction, Md is the reproductive output per original female during a period equal to d, and 0.74 is a correction factor.

Also, the net reproductive rates (R_0), the mean generation time (T), the doubling time (DT) and the finite rate of increase (λ) were estimated on each cultivar (Birch, 1948; South wood, 1978).

The demographic parameters were calculated in the basis of the following formulas:

1. The net reproductive rates, $R_0 = \sum l_x m_x$;

(l_x: The percentage of nymphs reached the adult stage, m_x : the daily fecundity of female).

2. The mean generation time, $T = \log R_0 / r_m$;
3. The doubling time, $DT = \log 2 / r_m$;
4. The finite rate of increase, (λ) = Total fecundity per female / total lifespan

Statistical analysis

The data concerning all biological and demographic parameters of the black bean aphid on five broad bean cultivars were compared using one-way analysis of variance (ANOVA). If the ANOVA demonstrated significant differences, the means were separated using the Tukey's test at $P \le 0.05$ (Gomez and Gomez, 1984). The nymphal mortality rate of *A. fabae* was subjected to the Chi-square test (χ^2) at 5% level. The experiment was arranged in the randomized complete block design (RCBD). A partial correlation model was adopted to determine the nature of the relationship between some biological parameters (weight of adult, size of adult, total number of embryos and number of developed embryos). All statistical analyses were performed with IBM SPSS statistical software (Version 23.0.0.0) (SPSS, 2015).

Results

Nymphal mortality rate of Aphis fabae

The results showed a significant difference ($P < 0.05$; $\chi^2_{9.48} = 10.91$) (Table 1) among the different cultivars. The cultivar 12 caused the highest mortality (26.83%), while the cultivar 4 had a very low effect on the mortality rate of nymphs (4.65%). The mortality rate of nymphs in the other cultivars was ranged from 11.36 to 25% (Table 1).

Table 1. Effects of five broad bean cultivars on the mortality rate of *Aphis fabae* nymph's.

Cultivars / Parameters	1	4	12	13	14	Total nymphs	Chi-square test
Nymphs reached the adult stage	32	41	30	30	39	172	χ^2 = 10.91
Dead nymphs	10	2	11	10	5	38	df = 4
Total nymphs	42	43	41	40	44	210	
Mortality rate (%)	23.81	04.65	26.83	25	11.36	18.10	P = 0.028*

*P < 0.05.

Size, weight and potential fecundity of females

The present experiment demonstrated significant difference in the adult size of the black bean aphid among the five broad bean cultivars tested ($F_{3.36}$ = 3.391; P = 0.049) (Fig. 1). The mean adult size was largest on the cultivar 1 (3.08 mm^2) and narrowest on the cultivar 12 (2.54 mm^2) (Fig. 1). Also, the data analysis indicated significant difference ($F_{3.18}$ = 3.260; P = 0.047) among the five broad bean cultivars in the adult weight of *A. fabae*. The heaviest adults were recorded on the cultivar 13 (1.18 mg), while the lightest aphids were found on the cultivar 12 (0.87 mg). The adult weight was ranged from 1.00 to 1.10 mg in the other cultivars (Fig. 2). In the present study, the effect of cultivars on total number of embryos was present ($F_{3.48}$ = 5.513; P = 0.013) (Fig. 3). The big total number of embryos was found on the cultivar 4 (29.33 embryos), while the small number was recorded on the cultivar 12 (20.33 embryos) (Fig. 3). Cultivars showed no significant effects on the number of developed embryos ($F_{3.48}$ = 2.474; P = 0.112). The mean number of developed embryos was ranged from 11.00 to 14.67 embryos in the five broad bean cultivars (Fig. 3).

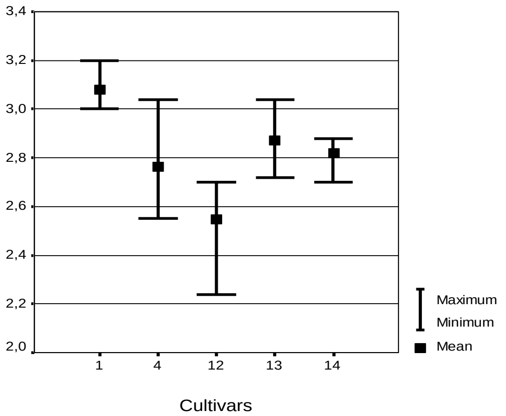

Fig. 1. Adult Sizes of *Aphis fabae* (mm^2) on five broad bean cultivars.

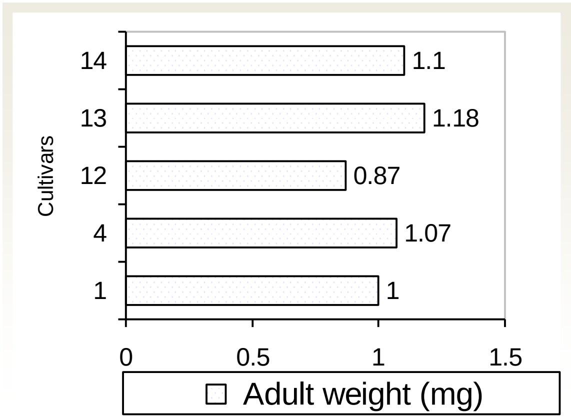

Fig. 2. Adult weights of *Aphis fabae* (mg) on five broad bean cultivars.

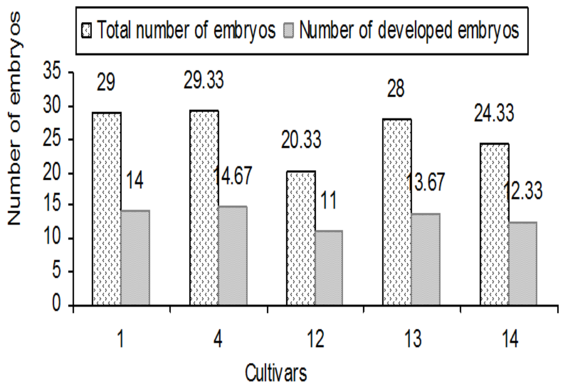

Fig. 3. Potential fecundity of females of *Aphis fabae* on five broad bean cultivars.

Relationship between biological parameters of Aphis fabae

The analysis of the partial correlation showed no relationship between all parameters except two parameters; total number of embryos and number of developed embryos, there is a positive low correlation between the two parameters (r = 0.568; P = 0.03) (Table 2).

Table 2. Correlation coefficients (r) between some biological parameters of *Aphis fabae* on five broad bean cultivars.

Parameters	Adult size	Adult weight	Total number of embryos	Number of developed embryos
Adult size	1.00			
Adult weight	0.229 (P = 0.431)	1.00		
Total number of embryos	0.323 (P = 0.260)	0.516 (P = 0.059)	1.00	
Number of developed embryos	0.341 (P = 0.232)	0.070 (P = 0.810)	0.568 (P = 0.03*)	1.00

* Significant at P ≤ 0.05.

Demographic parameters of Aphis fabae

The statistical analyses of all demographic parameters of *A. fabae*. Showed no significant differences among cultivars (P > 0.05) (Table 3).

Table 3. Summary of Black bean aphid Life table parameters on five broad bean cultivars (Means ± SE). n: number of repetitions, ns: not significant, within columns means followed by different letters are significantly different (ANOVA: P ≤ 0.05; Tukey's test).

Cultivars	Intrinsic rate of natural increase (r_m)	Net reproductive rate (R_0)	Generation time (T) (days)	Doubling time (days)	Finite rate of increase (λ)
1	0.217±0.012a	37.29±6.91a	16.63±1.11a	3.22±0.20a	1.87±0.29a
4	0.231±0.020a	65.70±6.14a	17.24±1.98a	3.06±0.27a	2.16±0.54a
12	0.228±0.007a	45.74±11.14a	16.55±0.91a	3.05±0.10a	2.66±0.81a
13	0.221±0.002a	56.80±16.88a	18.90±0.29a	3.14±0.03a	2.52±0.09a
14	0.236±0.005a	46.90±21.31a	1515±2.53a	2.94±0.06a	1.78±0.52
$F_{3.48}$	0.440	0.633	0.736	0.427	0.577
P	0.777 (ns)	0.650 (ns)	0.588 (ns)	0.786 (ns)	0.686 (ns)
n	3	3	3	3	3

Discussion

Understanding the biological and demographic parameters of a pest is essential to develop an integrated pest management strategy. These parameters provide population growth rate of an insect pest in the current and next generations (Frel *et al.,* 2003). The antibiosis category of plant resistance occurs when the negative effects of a resistant plant affect the biology of an arthropod attempting to use that plant as a host (Smith, 2005). The antibiotic effects of a resistant plant range from mild to lethal, and may result from both chemical and morphological plant defensive factors (Smith, 2005). The direct effects of antibiosis may also suffer the debilitating effects of reduced body size and weight, prolonged periods of development in the immature stages, and reduced fecundity (Smith, 2005).

To evaluate the resistance against plant pests, several parameters were used, among others, the size (Taylor, 1975), weight (Tolmay *et al.,* 1999), potential fecundity (Traicevski and Ward, 2002), duration of larval development (Kordan *et al.,* 2008), the longevity of adults (Faccoli, 2007), survival (Castro *et al.,* 2001), the daily fertility (Jimenez-Perez and Wang, 2004), the total fertility rate (Barre *et al.,* 2002), the intrinsic rate of natural increase (Laamari *et al.,* 2008), net reproductive rate (Marohasy, 1997) and doubling time (Sauge *et al.,* 1998), generation time (Kersting *et al.,* 1999) and the multiplication rate (Quénéhervé *et al.,* 2009).

There are generally two types of factors responsible for antibiotic resistance. Smith (2005) cited the allelochemicals as: toxins (alkaloids, ketones and organic acids), growth inhibitors (quercetin, oxalic acid, coumestrol and gossypol), physical and morphological barriers such as: plant structures; trichomes and the epicuticular lipids.

Berryman (1988) found that the factors of defense of plants against pests and pathogens may be constitutive or induced.

However, Norris (1986) mentioned that the power plant is commonly antibiotic depends on the quality and quantity of nutrients and allelochemicals of the host plant tissue. The same author reported that suitability or no suitability rather depends on the primary metabolites that are essential for normal development and reproduction of the insect.

Tolmay *et al.* (1999) noted that the reduction in insect size is a good parameter of antibiosis. The results of our experiment showed that the largest mean adult size of *A. fabae* was recorded on the cultivar 1 (3.08 mm^2), while the lowest value was reported on the cultivar 12 (2.54 mm^2). Thus Lebbal (2010) found that the largest adult size of *Aphis craccivora* was recorded on the cultivar 143 of bean.

However, Meradsi (2009) found that the adult size of *A. fabae* was not affected by the nutritional status of seven broad bean cultivars. The differences between the adult sizes of *A. fabae* were possibly related to differences in the content of the sap into amino acids. In aphids adult size depends on host age and species (Dixon, 1976), the level of amino-nitrogen (Chambers, 1979). An experiment on the rich phloem amino acids, demonstrated that the development of aphids was positively correlated with the highest values in asparagine and glutamine (Bernays and Chapman, 1994).

Chronic growth inhibition, due to either the presence of growth inhibitors or the absence of or reduction in the level of plant nutrients, is exhibited in several arthropod resistant cultivars (Smith, 2005). Annan *et al.* (1996) found that the ethyl acetate extracts of the leaves of some varieties of alfalfa had limited the growth of *A. craccivora*. Variations in aphid weight have often been attributed to changes in nutritional quality of host plants. For example, offspring born in low-quality environments are smaller at birth than offspring born in high-quality environments (Brough and Dixon, 1989).

The number of embryos obtained in females reared on cultivar 4 was almost 1.5 times higher than that counted in females reared on the cultivar 12.

Through this study, there was no correlation between all parameters tested ($P > 0.05$) except the total number of embryos and number of developed embryos. Between these last two parameters, a positive relationship exists ($r = 0.568$; $P = 0.03$). Indeed, Dixon and Dharma (1980) reported that the ovariole number is programmed and is not a function of adult weight or food quality for *A. fabae*. Concerning the weight at the black bean aphid, our results showed that the adults rearing on the cultivar 13 adults were heavier than those raised on the cultivar 12. There were a substances produced by the plant that stimulates the insect weight gain (Strebler, 1989). As adult weight increased so did offspring birth weight, regardless of the source host. This has been found for a number of aphid species such as *Rhopalosiphum padi* and *A. fabae* (Dixon, 1976, 1985). Time of birth also influenced birth weight: nymphs born to older mothers tended to be larger than those born earlier in the progeny sequence. This accord with the findings of Dixon and Wratten (1971) for *A. fabae*, Newton and Dixon (1990) for *Sitobion avenae*, and Dixon *et al.* (1993) for *Caveriella aegopodii* and *Megoura viciae*. Moreover, in the bird cherry-oat aphid, the number of embryos per ovariole was a constant, but the number of pigmented embryos present was a function of the size of the aphid at adult moult (Leather and Wellings, 1981). However, these same authors found that the number of embryos per ovariole in the sycamore aphid was dependent on the size of the adult. The differences in the biotic potential of individuals of *A. fabae* reared on the five broad bean cultivars probably assigned to a difference in the nutritional value or to the presence of substances that affects the fecundity.

The secondary metabolites had a key role in plant resistance against insects (Lachman *et al.*, 2001). Thus, the cultivar Radius of *Medicago sativa* resistant to *Acyrtosiphon pisum*, had a higher levels of saponins and a very low concentrations of flavonoids in comparison to the susceptible cultivar Sapko (Golawska *et al.*, 2008).

Organic acids in arthropod resistant plants have antibiotic effects (Smith, 2005). For example, DIMBOA was also an active component in the resistance of maize to the corn leaf aphid, *Rhopalosiphum maidis* (Long *et al.,* 1977). In aphids, the larviposition was closely related to the quality of the food preimaginal (Dixon, 1987). According to Mattson (1980), the dietary requirement of insects for nitrogen comprises both the total amount of nitrogen (nitrogen quantity) and the composition of nitrogenous compounds (nitrogen quality) of the food ingested. The phenolic compounds and their derivatives, such as quinones, tannins and lignins, were widely involved in plants resistance against pests (Strebler, 1989). The same author found that in addition to their impact on the behavior and specificity of plant-insect relationships, these compounds usually act on digestion and metabolism of insects.

Through this test, there were no significant differences between the five broad bean cultivars tested for all demographic parameters of *A. fabae*, among others the intrinsic rate of natural increase, the net reproductive rate, the doubling time, the generation time and the finite rate of increase. So, Meradsi (2009) found some differences between seven broad bean cultivars tested against *A. fabae* in parameters: the net reproductive rate, the generation time and the finite rate of increase. However, Laamari *et al.* (2008) found that the intrinsic rate of natural increase of *A. craccivora* was low (0.081) on the resistant broad bean cultivar V51, while it was high (0.116) on the susceptible cultivar V19.

This similarity of results of the demographic parameters may be related to no difference between the parameters used to calculate these parameters. As the pre-reproductive period and the daily fecundity. These two parameters were essential for calculating the intrinsic rate of natural increase and net reproductive rate. However, the generation time is closely linked by two parameters: the intrinsic rate of natural increase and net reproductive rate.

Thus, the calculated doubling time is attached by the intrinsic rate of natural increase. Probably for these reasons, there was no significant difference between five broad bean cultivars tested for the demographic parameters.

Conclusion

These preliminary data suggest that the mediocre performances of the black bean aphid were noted on the cultivar 12. But, the best performances of *A. fabae* were found on the susceptible control 4. This indicates that cultivar 12 or phytochemicals have potential as natural biopesticide for *A. fabae* control. The insecticidal effect could be related to the presence of different active compounds or a blend of toxics, which induce a high level of mortality.

The biological activity of the cultivar 12 should be further investigated under laboratory conditions to identify and evaluate specific biological and behavioral responses of *A. fabae*. This will help to identify precisely the main modes of action and levels of bioactivity of different compounds to better integrate them into management strategies for black bean aphid. Biological manipulation using natural products with fewer deleterious effects on non-targeted organisms and the environment for the management of aphids can be considered as a new approach for integrated pest management in field crops.

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