



Effect of the aphid rose *Macrosiphum rosae* (Hemiptera : Aphididae) infesting rose on physiological and natural characteristics of rose oil as well as population dynamics of this pest

Emam, A. S.; El-Roby, Afaf M. S. and Hassan, M. I.

Plant Protection Research Institute, Agricultural Research Centre, Dokki, Giza, Egypt.

ARTICLE INFO

Article History

Received: 18 / 4 / 2019

Accepted: 30 / 6 / 2019

Keywords

Rose, aphid rose, *Macrosiphum rosae*, rose oil, physiological and natural characteristics.

Abstract:

The aphid rose *Macrosiphum rosae* L. (Hemiptera : Aphididae) is stunts the growth of rose and reducing its market value. The aim of this research work is to study the population fluctuations of the aphid *M. rosae* during successive season 2018 on three varieties (colors) of rose and the effect of insect infestation by *M. rosae* on the physiological and natural characteristics of rose oil . The results indicated that the activity period for adults and nymphs of aphid rose infested the varieties Carmen, Golden gate and Dream were 23.5 adults/flower and 45.5 nymphs/flower; 25.6 adults/flower and 50.7 nymphs/flower and 20.5 adults/flower and 40.5 nymphs/flower on the mid of April, respectively. Also, this study was carried out to study effect of infested rose plants by *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil at El-Orman Garden (Giza Governorate) under glasshouse conditions during successive seasons 2018. Data obtained showed that the most important components of rose oil such as (geraniol, citronellol, nerol, stearpoten, phenyl ethanol and bioflavonoids), acids such as Citric acid and Malik acid and vitamins A, B, C and D were changed its concentrations as result of infestation by *M. rosae*. High infestation by *M. rosae* affected on concentrations of these components more than medium and low infestation compared to control. Also, data obtained showed that the most natural characteristics of rose oil such as volatility, light rotation and refraction value were changed as result of infestation by *M. rosae*, but other natural characteristics like freezing point did not change after infestation by *M. rosae*.

Introduction

Rose (*Rosa gallica*) is one of the most important ornamental plants in

Egypt and around all over the world which cultivated both in the open field

and under greenhouse conditions. So, it named king of flowers. It's found from oldest countries and it is the favorite flower for human in the world wide. Although developing live and highly technology but love human to roses still and increase. The human love for roses due to their beautiful colors, style of flowers, smiles and tolerant the inferable weather factors. Recently rose cultivated area increased gradually during the last years, especially in the new reclaimed areas for purposes local consumption and exportation to the foreign markets. So, rose became one of the important components for increase income for many countries all over the world, which producing and exporting these roses to different countries (Baydar ,2014 and Emam, 2009).

Rose plants infested with large scale of insects belong to many orders and families. *Macrosiphum rosae* L. (Hemiptera : Aphididae) commonly known as (Rose aphid) and is consider one of the most important insects of rose plants which infested both its leaves and flowers and also infested rose plants both in open fields and under green houses, Jaskiewicz (1997) who reported that the strong infestation by the rose aphid, *M. rosae* resulted in high deformation of stems, leaves and flowers of rose plants. Derek (2017) in Australia reported that *M. rosae* is a serious pest on rose and it is reproducing, parthenogenetically and viviparous all year round. It feeds mainly on the young leaves and developing flower-buds of roses. The adults and nymphs of aphid infest the rose plants and suck cell sap from flowers, tender shoots and buds, ultimately decreasing the market value of rose flowers, infestation with aphid causes badly affects the flowering capacity of plants about 20-40% losses. Labanowski (1989) in Poland reported that the rose aphid, *M. rosae* is the most important insect infests rose plants. Many authors dealt with the population dynamics of the rose aphid,

M. rosae i.e. Tomiuk and Wohrmann,1980 ; Rhomberg *et al.*, 1985; Ghosh *et al.*, 1994; Dixon, 1998 and Jaskiewicz, 2004.

The temperature does not only have a direct influence on the rose aphid but it also changes the physiology of the rose plant, which results in the stagnation of the plant. It would not be a suitable food source for the rose aphid. Therefore, aphids have to migrate to their secondary host plants to avoid these unsuitable conditions (Maelzer,1977 and Jaskiewicz ,1997).

This study was carried out to study effect of infested rose plants by *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil which found in rose flowers at El-Orman Garden, (Giza Governorate) under glasshouse conditions during successive seasons 2018. Therefore this study divided into two parts, first part study the population fluctuations of *M. rosae* during successive season 2018 on three varieties (colors) of rose and the second part included effect of insect infestation by *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil .

Materials and methods

1.Experimental design:

This study was conducted on three varieties (Carmen, Golden gate and Dream) of rose plants grown in El-Orman Garden, Giza Governorate under glasshouse conditions during successive seasons 2018. Two glasshouses with an area of 27x45 m of each one, was divided into 9 plots (3x5 m²), three plots for each variety of rose. The first one of these glasshouses contained infested rose plants and other one left as control. The first glasshouse was arranged in randomized block with three replicates to three varieties (colors) of rose and also the second glasshouse was arranged in randomized block with three replicates as control. The 1st glasshouse was

artificially infestation by *M. rosae* and the 2nd one was left as control. The two glasshouses were in an area isolated from other trees in the garden. Also, the first glasshouse was isolated from the second one. Rose plants were planted in glasshouse conditions at the same time on November (the planting time of rose plants). All agricultural operations of irrigation and fertilization and others are completely identical in the two glasshouses were done without application of any insecticide. Artificial infestation was done by the aphid *M. rosae* in the first glasshouse, with careful observation of the mean numbers of *M. rosae* during plant growth period and especially during the flowering stage from February – August and recorded mean numbers of aphid by direct counting biweekly, with examining the second glasshouse free as (control). With check up the physiological and natural characteristics of rose oil at three levels of infestations during three periods: winter during February month (medium infestation), spring during April month (high infestation) and summer during July month (low infestation).

2. Effect of insect infestation by *Macrosiphum rosae* on the physiological and natural characteristics of rose oil:

This study was carried out to study effect of infested rose plants by *M. rosae* on the physiological and natural characteristics of rose oil through determination the concentrations of most important components of rose oil such as (geraniol, citronellol, nerol, stearpoten, phenyl ethanol and bioflavonoids), acids such as Citric acid and Malik acid and vitamins such as A, B, C and D. Also, determination of the most important natural characteristics of rose oil such as volatility, light rotation, refraction value and freezing point.

3. Determination physiological and natural characteristics of rose oil:

3.1. Rose oil extraction:

Rose oil was extracted from 0.5 kg fresh tissue. The tissues were ground in liquid nitrogen with a mortar and pestle. Then few mls of tris buffer extraction were added (1:2, tissue: buffer). The medium of extraction contained tris-HCL buffer (0.1mM tris, pH 7.5, 4mM B-mercaptoethanol, 0.1mM EDTA-Na₂, 10mM KCl and 10mM MgCl₂). The crude homogenate was centrifuged at 10.000xg for 20Min. The supernatant was used for gel analysis by SDS-polyacrylamide gel electrophoresis (SDS-PAGE) according to the method of Laemmli (1970)

3.2. Loading on a gel:

3.2.1. Gel preparation:

Sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) was performed using 12.5% acrylamide and 0.8% bis acrylamide running gel consisting of 0.375 M Tris-HCl (pH 8.8) and 0.1% SDS. Stacking gel (10 mm) was made using 4.5% acrylamide containing 0.8% bis-acrylamide in 0.125 M Tris-HCl (pH6.8) and 0.1% SDS. The electrophoresis buffer contained 0.025 M Tris-HCl, 0.19 glycine and 0.1% SDS. The samples were homogenized in 0.12M Tris-HCl (pH 6.8), 0.4 SDS, 10 B-mercaptoethanol, 0.02% bromophenolbule and 20% glycerol. The samples were then heated for 3Min. in a boiling water bath before centrifugation. The gel was run under cooling at 90v for the first 15Min, then 120v the next 0.5 hr and finally 150v for the remaining 1.5hr (Sheri *et al.*, 2000).

3.2.2. Sample loading:

A known volume of protein sample was applied to each well by micropipette. Control wells were loaded with standard protein marker.

3.2.3. Electrophoresis conditions:

The running buffer was poured into pre-cooled (4°C) running tank. The

running buffer was added in the upper tank just before running, so that the gel was completely covered. The electrodes were connected to power supply adjusted at 100v until the bromophenol blue dye entered the resolving gel, and then increased to 250v until the bromophenol blue dye reaches the bottom of the resolving gel.

3.2.4. Gel Staining and destaining:

After the completion of the run, gel was placed in staining solution consisting of 1g of Coomassie Brilliant blue-R-250; 455 ml methanol; 90ml glacial acetic acid and completed to 1L with deionized distilled water. The gel was destained with 200ml destaining solution (100ml glacial acetic acid, 400ml methanol and completed to 1L by distilled water) and agitated gently on shaker. The destaining solution was changed several times until the gel background was clear.

3.2.5. Gel analysis:

Gels were photographed using a Bio-Rad gel documentation system. Data analysis was obtained by Bio-Rad Quantity one Software version 4.0.3, the sugar and protein were analyzed by High Pressure Liquid Chromatography (HPLC).

4. Statistical analysis:

In these experiments, effect of the infested rose plants by rose aphid *M. rosae* (in different stages of infestation) on the physiological and natural characteristics of rose oil was subjected to analysis of variance (ANOVA) and the means were compared by L.S.D. test at 0.05 level, using SAS program (SAS Institute, 1988).

Results and discussion

1. Population fluctuation of *Macrosiphum rosae* on rose flowers in Giza Governorate during 2018 season:

Data tabulated in Table (1) showed that the infestation by rose aphid, *M.*

rosae adults began to appear in Carmen variety on flowers on the 1st February with 6.5 adults/flower, then the infestation increased gradually to reach 23.5 adults/flower (activity peak) on the mid of April then the infestation decreased until reached to 1.7 adults/flower on mid of August. As the same trend nymphs began to appear in Carmen variety on the 1st February with 15.7 nymphs/flower, then the infestation increased gradually to reach 45.5 nymphs/flower (activity peak) on the mid of April then the infestation decreased until reached to 6.4 nymphs/flower on mid of August. Whereas for Golden gate variety *M. rosae* adults began to appear on flowers on the 1st February with 8.5 adults/flower, then the infestation increased gradually to reach 25.6 adults/flower (activity peak) on the mid of April then the infestation decreased until reached to 3.9 adults/flower on mid of August. As the same trend nymphs began to appear on flowers on the 1st February with 18.4 nymphs/flower, then the infestation increased gradually to reach 50.7 nymphs/flower (activity peak) on the mid of April then the infestation decreased until reached to 9.5 nymphs/flower on mid of August.

Also, for Dream variety *M. rosae* adults began to appear on flowers on the 1st February with 5.3 adults/flower, then the infestation increased gradually to reach 20.5 adults/flower (activity peak) on the mid of April then the infestation decreased until reached to 1.5 adults/flower on mid of August. As the same trend nymphs began to appear on flowers on the 1st February with 13.5 nymphs/flower, then the infestation increased gradually to reach 40.5 nymphs/flower (activity peak) on the mid of April then the infestation decreased until reached to 4.8 nymphs/flower on mid of August.

Table (1): Population fluctuation of *Macrosiphum rosae* (adults-nymphs) infested different rose varieties in Giza Governorate during 2018 season.

Date	Carmen (Red)		Golden gate (Yellow)		Dream (Pink)		Mean Temp.	Mean Hum. %
	Adult	Nymph	Adult	Nymph	Adult	Nymph		
1/2/2018	6.5	15.7	8.5	18.4	5.3	13.5	16.5	72
15/2/2018	12.8	21.8	15.3	25.9	10.5	18.6	18.3	73
1/3/2018	15.3	32.5	17.8	37.5	12.6	26.8	19.5	74
15/3/2018	18.5	37.7	19.3	40.5	15.4	32.5	18.4	70
1/4/2018	20.7	42.3	22.4	45.3	17.3	37.8	18.9	67
15/4/2018	23.5	45.5	25.6	50.7	20.5	40.5	22.7	63
1/5/2018	18.3	37.6	21.5	42.5	16.3	35.2	24.5	60
15/5/2018	15.2	30.5	18.3	36.7	12.3	26.7	25.2	53
1/6/2018	12.7	25.7	15.6	30.4	9.8	20.5	26.4	50
15/6/2018	10.5	20.8	13.4	25.7	7.2	16.7	27.1	47
1/7/2018	8.4	15.3	11.7	20.3	6.8	12.9	27.8	49
15/7/2018	5.9	11.6	8.4	15.4	3.6	9.6	28.6	53
1/8/2018	3.8	9.5	5.6	12.8	2.2	6.5	29.5	58
15/8/2018	1.7	6.4	3.9	9.5	1.5	4.8	30.7	56
Total	173.8	352.9	207.3	411.6	141.3	302.6	-	-
Mean	12.4	25.2	14.8	29.4	10.1	21.6	-	-
F(0.05)	475.85						-	-
L.S.D	1.97						-	-

Means within columns bearing different subscripts are significantly different ($P < 0.05$)

The obtained results are in agreement with those obtained by Mohammad and Al-Mallah (1987) in Iraq who stated that the first appearance of *M. rosae* in large numbers was in mid-February, peaked in early April and disappeared completely by mid-June. Jaskiewicz (2003) studied *M. rosae* population dynamics and found that maximum numbers of this insect was observed during March-May. Hole *et al.* (2017) studied incidence of the rose aphid, *M. rosae* on 30 rose varieties and found that the pest build up started on the third week of February which increased gradually reaching its peak on the fourth week of March and declined thereafter.

2. Effect of infested rose plants by rose

aphid, *Macrosiphum rosae* on the physiological and natural characteristics of rose oil:

2.1. Effect of infested rose plants by rose aphid, *Macrosiphum rosae* (in different stages of infestation) on the physiological characteristics of rose oil:

Data tabulated from Table (2) showed comparison between determinations of rose oil components (concentrations) in the rose flowers which infested by rose aphid, *M. rosae* in different stages of infestation (low, medium and high infestation) compared to control (non infested flowers). Data obtained showed that the most important components of rose oil (geraniol,

citronellol, nerol, stearpoten, phenyl ethanol and bioflavonoids), acids such as Citric acid and Malik acid and vitamins such as A, B, C and D were changed its concentrations after infestation by *M. rosae*. Concentrations of these components were more in control compared to its concentrations in

infested rose flowers (high, medium and low infestation, respectively). Statically analysis showed were highly significant differences between concentrations of these components in control compared to its concentrations in infested flowers (high, medium and low infestation), respectively.

Table (2): Determination of rose oil components (concentrations) on the different stages of infestation by rose aphid, *Macrosiphum rosae* in Giza Governorate during 2018 season.

Compounds	Concentrations of rose oil components (mg/g)				F(0.05)	L.S.D
	Low infestation	Medium infestation	High infestation	Control		
Geraniol	30.42 ^a	27.65 ^b	25.43 ^c	35.75 ^a	34.29***	1.18
Citronellol	20.18 ^b	17.35 ^b	15.45 ^c	25.25 ^a	25.31***	1.23
Nerol	16.45 ^c	14.21 ^b	12.54 ^c	18.35 ^a	19.45**	1.31
Stearpoten	31.45 ^b	27.34 ^b	23.56 ^c	35.21 ^a	23.71*	1.15
Phenyl ethanol	13.32 ^a	11.57 ^d	10.35 ^c	17.54 ^a	21.35**	0.75
Bioflavonoids	6.67 ^b	5.23 ^b	4.12 ^c	8.89 ^a	64.12**	0.63
Citric acid	10.21 ^b	8.67 ^b	6.25 ^c	12.23 ^a	34.18**	1.72
Malik acid	7.12 ^c	5.67 ^a	4.24 ^c	9.24 ^a	53.21***	0.16
Vitamin A	5.34 ^a	4.45 ^b	3.65 ^c	7.25 ^a	34.17*	0.08
Vitamin B	4.12 ^a	3.56 ^a	2.89 ^c	5.23 ^a	27.29***	1.15
Vitamin C	3.65 ^a	2.15 ^b	1.50 ^c	4.76 ^a	23.12**	0.37
Vitamin D	2.45 ^a	1.86 ^b	1.20 ^c	3.75 ^a	16.24*	1.42

Means within rows bearing different subscripts are significantly different (P < 0.05)

2.2. Effect of infested rose plants by rose aphid, *Macrosiphum rosae* (in different stages of infestation) on the natural characteristics of rose oil:

Data tabulated in Table (3) showed that comparison between natural characteristics of rose oil in rose flowers which infested by *M. rosae* in different stages of infestation (low, medium and high infestation) compared to control (non infested). Data showed that the most natural characteristics of rose oil

such as volatility, light rotation and refraction value were changed as result of infestation by *M. rosae*, but other natural characteristics such as freezing point did not change after infestation by *M. rosae*. Statically analysis show were highly significant differences between natural characteristics (except freezing point) of rose oil in infested rose flowers (high, medium and low infestation) compared to its natural characteristics in control.

Table (3): Natural characteristics of rose oil as result of infestation by rose aphid, *Macrosiphum rosae* in Giza governorate during 2018 season.

Characteristics	Low infestation	Medium infestation	High infestation	Control	F(0.05)	L.S.D
Volatility	0.812 ^a	0.765 ^b	0.760 ^c	0.855 ^a	33.97***	0.79
Light rotation	0.52 ^a	0.48 ^b	0.43 ^c	0.55 ^a	21.15**	0.58
Refraction value	1.420 ^a	1.400 ^b	1.365 ^c	1.460 ^a	31.28***	0.73
Freezing point	20C	20C	20C	20C	-	-

Means within rows bearing different subscripts are significantly different (P < 0.05)

The obtained results are in agreement with those obtained by Emam (2009) in Egypt who studied effect of infestation by *M. rosae* on the interior components of rose flowers, and found that natural characteristics of rose oil changed as result to the infestation by *M. rosae*. Peng and Miles (1991) studied the changes in the internal components of rose flowers such as rose oil, protein, sugar and vitamins, which infested with some insects and decided that the most effective in these components was the infestation by *M. rosae*. Becker and Apel (1992) reported that the decrease in concentrations of rose oil components may be due to the infestation by *M. rosae*. Atwal and Dhingra (2018) reported that the infestation by *M. rosae* was changed in the concentrations of rose oil components in the rose petals. While, Jaskiewicz (2006) studied the changes which happened in protein pattern in the rose petals which infested by *M. rosae*.

Also, the obtained results are in agreement with those obtained by Decheva (2015) in Bulgaria who investigated the changes in the rose oil components in flowers of rose plants, and found that the level of 12 rose oil components identified decreased as result of infestation by *M. rosae*.

References

- Atwal, A. S. and Dhingra, S. (2018):** Biological studies on *Macrosiphum rosae* the common rose aphid on rose plants. Indian J. Entomol., 33(2): 136-141.
- Baydar, H. (2014):** The cultivation and producing rose and rose oil industry in Turkey. Turktarm, 5(1): 54-57.
- Becker, W. and Apel, K. (1992):** Isolation and characterization of DNA clone encoding a novel – induced protein of rose oil. Plant Mol. Biol., 19: 1065-1067.
- Decheva, R.; Koseva, D. and Mikhailova, Y. (2015):** Some biochemical and histochemical changes in the tissues of the rose during and after dormancy. Fiziologiya na Rastenyata, 12(3): 76-84.
- Derek, A. (2017):** The biology and main causes of changes in number of the rose aphid, *Macrosiphum rosae* on cultivated roses in south Australia. Australian Journal of Zoology , 25(2): 269-284.
- Emam, A. S. (2009):** Effect of insect infestation on some rose plants. Ph.D. Thesis, Fac. Al-Azhar Universty., Cairo, Egypt.
- Ghosh, A. K.; Mier Durante, M. P. and Nieto Nafria, J. M. (1994):** Distribution of aphid fauna (Homoptera: Aphididae) in the north of Orocantabrian phytogeographic province, Spain. Boletin Asociacion Espanola Entomologia, 18(3–4): 81– 91.
- Hole, U. B.; Salunkhe, G. N. and Shirke, M. S. (2017):** Effect of meteorological parameters on population dynamics of rose aphid, *Macrosiphum rosae*. Annals of Plant Protection Sciences, 15(1):106-110.
- Jaskiewicz, B. (1997):** Observations on the occurrence on the rose aphid (*Macrosiphum rosae* L.) on bushes of *Rosa rugosa* Thunb. and *Rosa canina* L. Folia Horticulturae, 9 (1): 25-31.
- Jaskiewicz, B. (2003):** Population dynamics of *Macrosiphum rosae* inhabiting shrubs of *Rosa* sp. In the green areas of Lublin. Annales universitatis Mariae Curie EEE, Horticultura, 13: 271-278.
- Jaśkiewicz, B. (2004):** The population dynamics of aphids colonizing the shrubs of *rosa canina* L in Urban conditions of Lublin, EJPAU 7(2): #05.Available
Online: <http://www.ejpau.media.pl/volume7/issue2/horticulture/abs-05.html>

- Jaskiewicz, B. (2006):** The effect of the feeding of *Macrosiphum rosae* on the flowering of roses. Acta-Agrobotanica, 59(1): 515-520.
- Labanowski, G. (1989):** Pests of ornamental plants: The Rose aphid, *Macrosiphum rosae*. Ochrona Roslin, 4(2): 33-39.
- Laemmler, U. K. (1970):** Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature, 15(27): 680 – 685.
- Maelzer, D. A. (1977):** The biology and main causes of changes in numbers of the rose aphid, *Macrosiphum rosae* (L.), on cultivated roses in South Australia. Australian Journal of Zoology, 25: 269– 284.
- Mohammad, M. A. and Al-Mallah, N. M. (1987):** Ecological and biological studies on the Rose aphid, *Macrosiphum rosae* in Mosul region. Arab J. Plant Prot., 5(2): 53-58.
- Peng, Z. and Miles, P. (1991):** Oxidases in the gut of an aphid *Macrosiphum rosae* (L.) and their relation to dietary phenolics. Journal of Insect Physiology, 37(10): 779-787.
- Rhomberg, L. R.; Joseph, S. and Singh, R. S. (1985):** Seasonal variation and clonal selection in cyclically parthenogenetic rose aphids (*Macrosiphum rosae*). Canadian Journal of Genetics and Cytology, 27(2): 224–232.
- SAS Institute (1988):** SAS/STAT User Guide, Ver. 6. 03. SAS Institute Inc., Cary, North Carolina.
- Sheri, L. H.; Necolas, E. S. and Joanna, B. G. (2000):** Comparison of protein expressed by *Pseudomonas aeruginosa* strains representing initial and chronic isolated from a cystic fibrosis: an analysis by 2-D gel electrophoresis and capillary liquid chromatography tandem mass spectrometry. Microbiol, 146: 2495-2508.
- Tomiuk, J. and Woehrmann, K. (1980):** Population growth and population structure of natural populations of *Macrosiphum rosae* (L.) (Hemiptera: Aphididae). Zeitschrift fur Angewandte Entomologie, 90:464 –473.