

## Effect of Nitrogen Fertilizer on Biological Parameters of the *Aphis craccivora* (Hemiptera: Aphidiae) and Associated Productivity Losses in Common Globe Amaranth

A. Hosseini<sup>1</sup>, M. Hosseini<sup>1\*</sup>, M. Goldani<sup>2</sup>, J. Karimi<sup>1</sup>, and H. Madadi<sup>3</sup>

### ABSTRACT

Reducing nitrogen status of floriculture plants is an effective tactic in pest management by decreasing host plant quality. Life table parameters and population growth rate of *Aphis craccivora* (Hemiptera: Aphidiae), as well as cosmetic and qualitative parameters losses associated with aphid infestation were evaluated in relation to N fertilization levels on *Gomphrena globosa*, under greenhouse condition. Four N fertilization levels (0, 30, 60, and 100% of recommended 2 kg m<sup>-3</sup>) were used. The results indicated that aphid's intrinsic rate of natural increase on plants fertilized with 100% of the recommended N level was the highest. Abundance and population growth rate of aphid also positively correlated with N fertilization levels. The interactive effect of aphid population and N fertility significantly affected growth parameters of the plants. In the absence of aphid, plant yield improved linearly with increasing N levels. However, aphid population highly decreased shoot to root ratio and the number of flowers in plants fertilized with 100% of the recommended N level. According to our findings, fine-tuning fertility to reduce *A. craccivora* population is a steadfast tactic to produce marketable globe amaranth ornamental plant.

**Keywords:** Aphid performance, *Aphis craccivora*, Nitrogen fertilization, Plant yield, Population growth.

### INTRODUCTION

*Gomphrena globosa* L. (Amarantaceae), the common globe amaranth, is a popular ornamental plant grown as cut flower, annual beds, border and potted flowering plants in mild-climate regions (Dinda *et al.*, 2006). This ornamental plant has relatively few pests (Bachman, 2011), among which the cowpea aphid, *Aphis craccivora* Koch (Hemiptera: Aphididae), is a serious one (Lovisollo and Conti, 1966; Jones, 1967; Tao, 1990). This aphid causes plant growth stunting, deformed leaves, and delay in

initiation of flowering by feeding phloem sap and transmission of viral diseases. Even in heavy attack, it can cause plant death (Obopile, 2006). In consequence, cosmetic parameters of ornamental plants such as *G. globosa* devalue and crop yield reduction (Byrne *et al.*, 1990; Alfonsina, 2008).

Nutritional quality of plant plays a significant role in host plant-herbivore interactions (Mattson, 1980; White, 1993; Trdan *et al.*, 2005; Tradan *et al.*, 2008). The most critical macronutrient, which profoundly influences the growth and fecundity of herbivorous insects, is nitrogen (Mc Neill and Southwood, 1978; Douglas,

<sup>1</sup> Department of Plant Protection, College of Agriculture, Ferdowsi University of Mashhad, Mashhad, Islamic Republic of Iran.

\* Corresponding author; email : m.hosseini@um.ac.ir

<sup>2</sup> Department of Agronomy, College of Agriculture, Ferdowsi University of Mashhad, Mashhad, Islamic Republic of Iran.

<sup>3</sup> Department of Entomology, Faculty of Agriculture, Bu Ali Sina University, Hamedan, Islamic Republic of Iran.



2006). Probably, one important reason for this phenomenon is the vast differences in N compound concentration between herbivorous insects (6-10%) and plant tissues (1.5–3%) (Mattson, 1980; Elser *et al.*, 2000). Scarcity of N compounds is especially true for phloem-feeding insects (e.g. aphids) because of the low N content of phloem (0.004-0.06%) compared to other plant tissues (Minkenberg and Fredrix, 1989; Ponder *et al.*, 2000). One of the cultural practices of ornamental plants that is commonly applied to improve their commercial and aesthetic aspects is N fertilization (N) (Eliott *et al.*, 2004; Chau *et al.*, 2005). However, N fertility, with increasing amino acid and nitrate level in host plant (Mengel and Kirkby, 2001) can enhance nutritional quality and attractiveness of plants for herbivorous insects; therefore, it improves performance parameters of phloem feeders (Mattson, 1980; White, 1993; Douglas, 2006; Fallahpour *et al.*, 2015). For instance, results of a study conducted by Aqueel and Leather (2011) showed that 0.4 g plant<sup>-1</sup> N fertilizer in the form of ammonium nitrate significantly enhanced the fecundity and longevity of *Sitobion avenae* (F.) and *Rhopalosiphum padi* (L.) in comparison to those by 0.1 g plant<sup>-1</sup> N fertilizer level. Zehnder and Hunter (2008) also found an increase in per capita population growth of *Aphis nerii* Boyer de Fonscolombe on *Asclepias tuberosa* L., i.e. plants with application of N fertilization in the form of ammonium nitrate.

Thus, based on previous reports, lowering N fertilization level, which may reduce yield and quality of ornamental plants a little, could be a useful management tactic to decrease pest population and control costs (Chau *et al.*, 2005; Chow *et al.*, 2011). In the north and northeast of Iran, *G. globosa* plants have been produced as a popular floriculture crop (Ghasemi and Kafi, 2007). The results of earlier studies indicated that the nursery of this plant needed a high N fertilization regime due to its fast-growing and highly-

demanding nutrition (Huang *et al.*, 2002; Benary, 2011) so as to increase their susceptibility to damage caused by aphid infestation. Thus, N fertilizer usage should be optimized to a level that markedly maintains physiological status of plant optimum and aesthetic parameters whilst minimizing the aphid population growth.

In this study, the specific objectives were to: (1) Assess the effect of N fertilization levels on life table parameters of the cowpea aphid on *G. globosa* plants, (2) Evaluate the effect of N fertilization on aphid population growth rate and the associated amaranth plants productivity (e.g. number of flower buds) and quality losses (e.g. nitrogen-compounds status of leaf) under greenhouse conditions.

## MATERIALS AND METHODS

### Host Plant, Fertilization and Aphid Population

Sixty four seedlings (7 cm in height) of the common globe amaranth were transplanted into separated plastic pots (23×17 cm, height×diameter) on July 5<sup>th</sup>, 2011. The pots were filled with 3 kg substrate including perlite (Mehr Parsian Exir Company, Iran), sphagnum peat moss (Kuomari Coir Products, Singapore), and sandy soil (ratio 1:1:1 v/v) as a growing media (Huang *et al.*, 2002). The seedlings were irrigated two times daily for 4 weeks with distilled water. One month after transplanting, the seedlings (17 cm in height) were subsequently assigned to different N treatments until the end of experiments. The recommended N level for the periodic feeding (thrice weekly for two months) of potted amaranth plants is approximately 2 kg m<sup>-3</sup> (as slow release N fertilizer) (Benary, 2011). The volume of amaranth plant pot was 7.6×10<sup>-3</sup> m<sup>3</sup>, so the recommended level of N fertilization per pot was calculated at 12 g. Based on N fertilizer (Urea, CO (NH<sub>2</sub>)<sub>2</sub> as source of nitrogen), four different N treatments including 0, 30,

60, and 100% of the recommended N level (12 g), were applied. The amounts of phosphorous (P) (trisodium phosphate as source of phosphorous) and potassium (K) (potassium sulfate as source of potassium) in all N fertilization treatments were equal to 10 g (52 g trisodium phosphate) and 20 g (44 g potassium sulfate), respectively. These amounts were separately given to the units three times for two weeks, starting with N application. The plants received recommended nutrient (N, P, K) solution (100 ml), and, in the intervals between fertilizers applications, were tap watered. All plants were kept at  $26\pm 1^\circ\text{C}$ ,  $65\pm 5\%$  RH and a light: dark regime of 16 L:8 D hour. The study was terminated on October 30<sup>th</sup>, 2011.

Aphids used in this study were procured from an *A. craccivora* colony, originally established with individuals collected from bean, *Phaseolus vulgaris* L., grown in a research greenhouse at the College of Agriculture of Ferdowsi University of Mashhad, Iran. The aphids were grown on vegetative *G. globosa* plants within net-covered cages (60×70 cm, height×diameter) for 53 days. The cages were kept in a growth chamber at  $26\pm 1^\circ\text{C}$ ,  $65\pm 5\%$  RH and a light: dark regime of 16 L:8 D hour.

#### Effects of N fertilization Levels on Life Table Parameters of *A. craccivora*

To determine the performance of *A. craccivora* on amaranth plants fertilized with different N regimes, life table parameters of aphid were considered as trial 1. Eight amaranth plants were assigned to each of the four N fertilization treatments. In each N fertilization treatment, 35 adult aphids from the reared colony were placed individually on the surface of young expanded plant leaves (90 days old) using camel's hair brush. A ventilated clip cage (1.5 cm<sup>2</sup> in diameter) engaged each individual aphid. After 24 hours, adults and all the youngs, except one first instar, were removed. This single aphid was allowed to develop to adulthood. All aphids were

checked daily from the onset of reproduction and every other day thereafter. The number of young nymphs produced by each aphid (age-specific fecundity) was also recorded daily and then removed during the remainder of its life. This trial was done in a growth chamber at  $26\pm 1^\circ\text{C}$ ,  $65\pm 5\%$  RH, and 16:8 hour (L: D) photoperiod. To construct the age-specific fertility life table, age specific survival rate ( $l_x$ ) and average aphids progeny in  $x$  age class ( $m_x$ ) were obtained. Based on these data, the intrinsic rate of natural increase ( $r_m$ , female progeny per female per day) was estimated using the following equation (Birch, 1948):

$$\sum L_x m_x e^{-r_m \text{pivotal} x} = 1 \quad (1)$$

Where,  $L_x$  and *pivotal*  $x$  are  $(l_x + l_{x+1})/2$  and  $[x + (x+1)/2]$ , respectively. Other parameters of fertility life table including net reproduction rate ( $R_0 = \sum l_x m_x$ ), Doubling Time ( $DT = (\ln 2)/r_m$ ), mean generation time ( $T = (\ln R_0)/r_m$ ) and finite rate of increase ( $\lambda = e^{r_m}$ ) were likewise computed (Carey, 1993). To find the differences in  $R_0$ ,  $T$ ,  $DT$ ,  $\lambda$  and  $r_m$  values, Jackknife method was applied for producing *pseudo*-values (Meyer *et al.*, 1986; Maia *et al.*, 2000).

To assess the impact of different N fertilization levels on the aphid performance, the life table parameters of *A. craccivora*, except aphid nymphal survivorship which was analyzed with non-parametric Kruskal-Wallis test, were subjected to one-way ANOVA (SAS Institute Inc. 2003). Accordingly, if significant differences between means were detected ( $P \leq 0.05$ ), Fisher's Protected *LSD* test was used. Before ANOVA, the normality of the data was tested by Kolmogrov-Smirnov in Minitab software (Minitab 16 statistical software). Assumption of the homogeneity of data was tested through Bartlett's test in the same software.

#### Effect of N Fertilization Levels on Aphid Population Growth Rate



This experiment was a completely randomized design (4×2 full factorial) with the four levels of fertilization (0, 30, 60, and 100% of the recommended N level) crossed with two levels of aphid densities (12 individuals of *A. craccivora* or no aphids) as trial 2. The experimental unit consisted of an individual amaranth plant enclosed in a 70×30 cm (height×diameter), cylindrical-shaped clear plastic container topped with an organdy-mesh cover. Eight units were used for each of the four N fertilization treatments. Half of the units per treatment were randomly selected and inoculated with aphids when the plants were 13 weeks old (60 days after the plants were assigned to the treatments). To approach a stable age distribution and allow the population to grow exponentially from the start of the experiment (Vehrs *et al.* 1992), variously-aged aphids (four adults, 4 L 3-4, 4 L 1-2) were transferred, using a camel's hair brush, to the apical region of the plant in each unit. To assess the effect of aphid infestation on yield losses and cosmetic parameters of amaranth plants, the other 4 units were kept free of aphids. Aphid infested (AI) and non-infested (non-AI) plants were grown in two growth chambers (26±2°C, 65±10% RH, 16 L: 8 D hour) to avoid aphid's contamination. Both growth chambers had the same environmental conditions.

Fifteen days later, equivalent to approximate 4×(multiplication) the doubling time of *A. craccivora*, the total number of aphids was counted visually in each unit of N treatments and population growth rate ( $r$ ) of aphid was estimated using the following formula (Hosseini *et al.*, 2010a):

$$r = \frac{\ln(N_{x+1} / N_x)}{t} \quad (2)$$

Where,  $N_x$  is the population size at time  $x$ ,  $N_{x+1}$  the population size at time  $x+1$  and  $t$  the difference in days between time  $x+1$  and  $x$ .

One-way ANOVA was applied to analyze the effect of fertilization levels on the overall population growth rate of the aphid.

Whenever a significant result for the ANOVA was obtained ( $P \leq 0.05$ ), Fisher's Protected *LSD* test was performed to determine the significance of differences between means.

### Effect of N Fertilization Levels on *G. globosa* Growth Parameters

At the termination of trial 2, net photosynthesis, stomatal conductivity, leaf chlorophyll content, leaf area, flower and bud number, and plant height as well as dry weight of shoot (total plant top), root, leaf and central stem of amaranth plant were measured to characterize yield of plant in AI and non-AI units.

Net photosynthesis rate ( $P_n$ ) and stomatal conductivity ( $g_s$ ) were taken between 8:00 and 9:00 a.m. in each fertilization treatment using portable photosynthesis system (LI-COR Inc., Lincoln, NE, USA) and leaf porometer system (Sc-1), respectively. In both measurements, three new expanded leaves from each pot (either AI or non-AI pot) were randomly selected. There were 6 replications per treatment combination ( $n=6$ ).

Leaf chlorophyll content was obtained with a SPAD portable leaf chlorophyll meter (SPAD-502, Minolta Camera, Co. Japan). Each leaf was one replication, and there were three replications per treatment combination ( $n=3$ ). During the study,  $P_n$ ,  $g_s$  and leaf chlorophyll content were measured once a week, but only the last data were reported.

For measuring shoot dry matter (equal to leaf and central stem DM) and root in each pot, total top plant (whole aerial shoot of the plant) and root were harvested and lightly washed with deionized water. Then, the leaves, central stem and root of harvested plants were detached and separately oven-dried for 48 hours at 70°C and weighed by a digital balance (Sartorius GD503, Germany, sensitivity 1mg) to obtain shoot dry matter.

The acquired data was analyzed using a two-way factorial ANOVA to assess the

effect of interaction between N fertilization levels and aphid presence. Significant differences were determined ( $P \leq 0.05$ ) by Fisher's *LSD* test. To estimate the effect of N fertilization level on population growth rate of *A. craccivora* and the associated losses of flower number and shoot to root ratio in amaranth plant, the linear regression model was used.

### Effect of N Fertilization Levels on *G. globosa* Nutrient Quality

At the termination of trial 2, to measure values of leaves total N and carbon contents, apical leaves (each leaf was a single replication) were dried like previous experiment and packed into tin capsules (each sample containing 2-3 mg dry matter). For each element (N and C), 6 replications per treatment were used. The total N and C contents were measured using the Kjeldahl procedure (Jones, 1984) and simple acidimetric titration (Richards, 1954), respectively.

Nutritional variables were subjected to two-way ANOVA to evaluate the interactive effect of N fertilization levels and aphid presence. The means were separated by the Least Significant Difference (LSD) test when a significant F-value ( $P \leq 0.05$ ) was obtained.

**Table 1.** Life table parameters (mean $\pm$ SE) of *Aphis craccivora* on *Gomphrena globosa* plants grown under different N levels (n= 35).<sup>a</sup>

Parameters <sup>b</sup>	Nitrogen treatment (% of recommended N level)				LSD
	0	30	60	100	
Juvenile survivorship	18 a	19 a	24 a	25 a	-
Developmental time (Days)	9.4 $\pm$ 0.11 a	9.6 $\pm$ 0.23 a	9.5 $\pm$ 0.13 a	9.7 $\pm$ 0.18 a	0.528
Adult longevity (Days)	9.3 $\pm$ 1.75 a	12 $\pm$ 1.65 a	11.2 $\pm$ 1.58 a	13.4 $\pm$ 1.11 a	4.675
Total nymph per adult	7.9 $\pm$ 1.2 c	9.2 $\pm$ 1.2 b	9.6 $\pm$ 0.9 b	12.4 $\pm$ 1.1 a	3.303
R <sub>0</sub> (Female/Female/Generation)	4.9 $\pm$ 0.92 c	7.4 $\pm$ 0.94 b	8.7 $\pm$ 0.89 b	12.2 $\pm$ 1.05 a	2.856
r <sub>m</sub> (Female/Female/Day)	0.12 $\pm$ 0.007 c	0.147 $\pm$ 0.005 b	0.16 $\pm$ 0.006 b	0.178 $\pm$ 0.006a	0.024
λ (Female/Female/Day)	1.12 $\pm$ 0.01 c	1.15 $\pm$ 0.01 b	1.17 $\pm$ 0.007 b	1.2 $\pm$ 0.007 a	0.023
T <sub>c</sub> (Days)	13.62 $\pm$ 0.27 a	13.74 $\pm$ 0.29 a	13.46 $\pm$ 0.27 a	14 $\pm$ 0.240a	0.811
D <sub>t</sub> (Days)	5.51 $\pm$ 0.253 a	4.65 $\pm$ 0.15 b	4.35 $\pm$ 0.162 b	3.85 $\pm$ 0.16 c	0.554

<sup>a</sup> Within the same row, means followed by the same letters indicate that they were not significantly different ( $P = 0.05$ , LSD after one-way ANOVA). <sup>b</sup> r<sub>m</sub>: Intrinsic rate of increase; R<sub>0</sub>: Net reproductive rate; λ: Finite rate of increase; T<sub>c</sub>: Mean generation time (days), and D<sub>t</sub>: Doubling time (days).

## RESULTS

### Effects of N Fertilization Levels on Life Table Parameters of *A. craccivora*

#### Aphid nymphal Developmental Time and Survivorship

Nymphal developmental time of *Aphis craccivora* was not influenced by N fertilization levels ( $F_{3, 79} = 0.7$ ,  $P > 0.05$ , Table 1). Likewise, no significant N treatment effect was observed for aphid juvenile survivorship ( $H = 1.6$ ,  $P > 0.05$ , Table 1).

#### Aphid Adult Longevity and Reproduction

The applied N fertilization levels did not have any significant effect on aphid adult longevity ( $F_{3, 76} = 1.08$ ,  $P > 0.05$ , Table 1). The highest number of offspring per adult aphid (12.4 $\pm$ 1.1) was acquired on the plants fertilized with 100% recommended N level and the lowest number (7.9 $\pm$ 1.2) was observed on the plants not fertilized ( $F_{3, 76} = 2.68$ ,  $P < 0.05$ , Table 1).

#### Age-Specific Fertility Life Table Parameters

Aphids feeding on the plants that received the greatest N level (12 g) had the highest r<sub>m</sub> and the lowest value was observed in aphid



reared on non-N-fertilized plants (0) ( $F_{3,74}=2.48$ ,  $P < 0.05$ , Table 1). Likewise, the net reproductive rate ( $R_0$ ) and finite rate of increase ( $\lambda$ ) of *A. craccivora* positively grew by increasing N fertilization levels ( $R_0$ :  $F_{3,74}=9.31$ ,  $P < 0.05$ ;  $\lambda$ :  $F_{3,74}=11.87$ ,  $P < 0.05$ , Table 1). Aphids reared on plants with 100% recommended N level had the shortest doubling time ( $D_T$ :  $F_{3,74}=13.64$ ,  $P < 0.05$ ). However, N fertilization levels did not have any significant effect on aphid generation time (T) ( $T$ :  $F_{3,74}=0.72$ ,  $P > 0.05$ , Table 1).

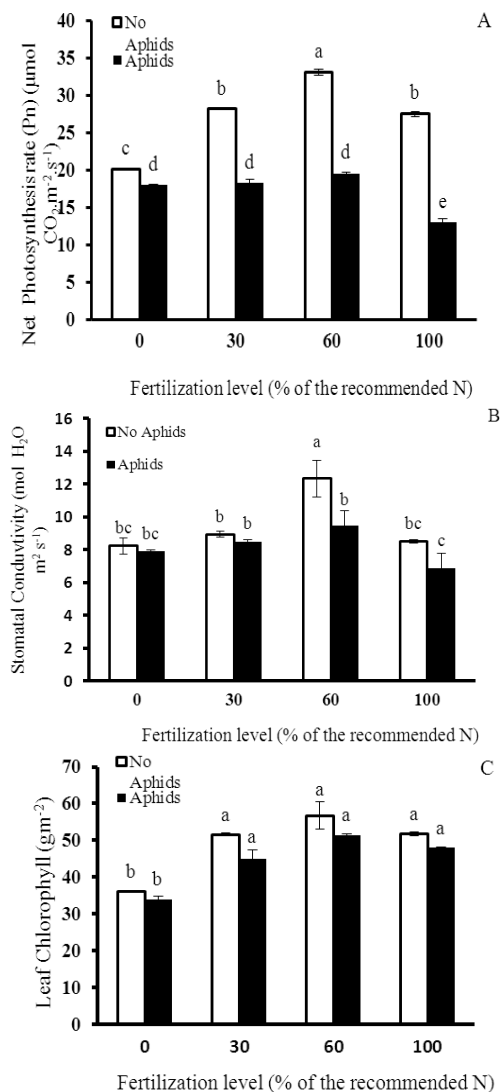
### Effect of N Fertilization Levels on Aphid Population and Quality Losses of *G. globosa* Plants

Results of linear regression analysis revealed that the growth population rate ( $r$ ) of *A. craccivora* grew along with increasing N fertilization levels to reach a maximum level ( $0.141 \pm 0.006$ ) for aphids reared on amaranth plants fertilized with 100% recommended N level ( $F_{3,12}=6.05$ ,  $P < 0.05$ ,  $Y=0.004X+0.9$ ,  $P < 0.05$ ,  $R^2=0.58$ ).

*A. craccivora* infestation significantly reduced both vegetative and reproductive growth parameters of *AI* plants.  $P_n$  and  $g_s$  were the highest in *non-AI* plants, fertilized with 60% of the recommended N level, while they were lowest in *AI* plants fertilized with 100% recommended N [ $P_n$ :  $F_{3,47}=8.19$ ,  $P < 0.05$ ,  $g_s$ :  $F_{3,47}=5.85$ ,  $P < 0.05$ ; Figure 1 (A and B)].

There was a significant interactive effect of N fertilization levels and aphid presence on chlorophyll content of leaves ( $F_{3,23}=3.68$ ,  $P < 0.05$ ). Chlorophyll content did not have a significant difference among N fertilized plants (Figure 1-C). However, in non-fertilized treatment, there was a significant fall in chlorophyll content of both *AI* and *non-AI* plants.

In the 100% N recommended fertilized *non-AI* plants, root Dry Matter (DM) was markedly lower than that in other treatment combinations ( $F_{3,23}=9.76$ ,  $P < 0.01$ , Table 2). In the highest N (100%) received *non-AI* plants, central stem DM increased



**Figure 1.** Effect of N fertilization levels and aphid presence on: (A) Leaf chlorophyll content [effect of N fertilization, aphid presence and both of them on leaf chlorophyll content were respectively,  $P < 0.05$ , no significant and  $P < 0.05$ ]; (B) leaf stomatal conductivity ( $g_s$ ) [effect of N fertilization, aphid presence and both of them on leaf  $g_s$  were respectively,  $P < 0.05$ ,  $P < 0.05$  and  $P < 0.05$ ]; and (C) Net photosynthesis rate ( $P_n$ ) [effect of N fertilization, aphid presence and both of them on  $P_n$  were respectively,  $P < 0.05$ ,  $P < 0.05$  and  $P < 0.05$ ].  $n=3$  for leaf chlorophyll;  $n=6$  for  $P_n$  and  $g_s$ ; bars indicate the standard error of mean, when absent it falls under the symbol, and means with different letters indicate significant differences among treatment combinations ( $P < 0.05$ , LSD after two-way factorial ANOVA).

**Table 2.** Effect of N fertilization levels and *Aphis craccivora* abundance on Carbon: Nitrogen ratio (C:N), Nitrogen (N) content (%), Carbon (C) content (%), Shoot: Root ratio [Dry Matter (g)= DM], Number (No.) of flower, root DM, leaf DM, central stem DM (mean±SE).<sup>a</sup>

Parameters	Nitrogen treatments (% of recommended N level)							
	0		30		60		100	
	Aphid	No aphid	Aphid	No aphid	Aphid	No aphid	Aphid	No aphid
C:N	1.43 ± 0.04b	2.3 ± 0.25a	0.65 ± 0.11c	0.98 ± 0.45bc	0.68 ± 0.1c	0.73 ± 0.06c	0.60 ± 0.03c	0.74 ± 0.013c
N content	0.8 ± 0.08c	0.87 ± 0.041c	1.14 ± 0.09b	1.31 ± 0.072b	1.46 ± 0.14b	1.82 ± 0.0a	1.45 ± 0.16b	2.15 ± 0.041a
C content	1.85 ± 0.12a	1.97 ± 0.05ab	1.39 ± 0.02de	1.69 ± 0.005bc	1.7 ± 0.045bc	1.3 ± 0.08ef	1.13 ± 0.05dc	1.57 ± 0.07f
Shoot: Root	5.5 ± 0.94d	4.8 ± 0.65d	7.17 ± 0.57cd	9.45 ± 0.28bc	8.94 ± 0.67c	10.94 ± 0.57b	6.6 ± 0.54cd	16.45 ± 0.05a
No of flower	41 ± 0.28d	45 ± 3.46d	57.5 ± 9.4c	68 ± 6.9bc	70.25 ± 6.8bc	74 ± 2.45b	75 ± 0.58b	96 ± 0.58a
Root DM	2.72 ± 0.15a	3.05 ± 0.28a	2.5 ± 0.15a	2.6 ± 0.28a	2.47 ± 0.10a	3.65 ± 0.37a	2.47 ± 0.03a	0.98 ± 0.02b
Leaf DM	0.05 ± 0c	0.04 ± 0.001c	0.09 ± 0.008b	0.08 ± 0.008b	0.11 ± 0.005ab	0.12 ± 0.007a	0.11 ± 0.002ab	0.09 ± 0.001b
Central stem DM	8.5 ± 0.15c	9 ± 0.404bc	9.36 ± 0.12b	9.56 ± 0.09b	11.21 ± 0.2ab	12.2 ± 1.5a	9.55 ± 0.1b	13.12 ± 1.2a

<sup>a</sup> P= 0.05, LSD after two-way ANOVA.

significantly ( $F_{1, 23} = 5.03$ ,  $P < 0.05$ , Table 2). Leaf DM in *non-AI* plants fed either with 60 or 100% N reached the highest level ( $F_{1, 23} = 52.70$ ,  $P < 0.01$ , Table 2). The number of flowers and shoot to root ratio were significantly different in treatment combinations (Number of flower:  $F_{3, 23} = 16.06$ ,  $P < 0.01$ ; Shoot to root ratio:  $F_{3, 23} = 48.02$ ,  $P < 0.01$ ), and those were highest in *non-AI* plants fertilized with 100% N recommended level (Table 2).

The regression analyses confirmed a major positive relationship between N fertilization level and both the number of flowers and shoot to root ratio in the absence of aphid [Shoot to root ratio (A):  $Y = 0.11X + 5.4$ ,  $P < 0.01$ ,  $R^2 = 0.75$ ; Number of flowers (B):  $Y = 0.48X + 47.9$ ,  $P < 0.01$ ,  $R^2 = 0.77$ ]. Nevertheless, in the presence of aphid, these relationships were less strong than in the absence of aphid, both regarding shoot to root ratio ( $Y = 0.02X + 6.4$ ,  $P < 0.05$ ,  $R^2 = 0.45$ ) and the number of flowers ( $Y = 0.31X + 44.9$ ,  $P < 0.01$ ,  $R^2 = 0.88$ ).

Leaf-N content was at its peak in *non-AI* plants fertilized with either 60% or 100% of the recommended N level in comparison with the other treatment combinations (N:  $F_{1,35} = 6.89$ ,  $P < 0.05$ ; C/N:  $F_{1,35} = 11.76$ ,  $P < 0.05$ , Table 2). In addition, in non-fertilized plants, leaf-N content markedly decreased. The highest C/N ratio was obtained in *non-AI* plants with no fertility; whereas the lowest C/N ratio was found in both *non-AI* and *AI* plants fertilized with either 60% or 100% of the recommended N level (Table 2).

## DISCUSSION

Based on evidence in the literature, N fertilization of ornamental plants generally leads to increased growth rate and higher potential fecundity of insect herbivores and, ultimately, to increased population densities (Bentz and Larew, 1992; Davies *et al.*, 2004; Hogendorp *et al.*, 2006; Chow *et al.*, 2011). The results of the present study also confirmed that some life table parameters, such as the



intrinsic rate of increase ( $r_m$ ) of *A. craccivora* positively correlated with the application of N fertilization in *G. globosa* plants. Nevertheless, since there was not a significant difference of developmental time of *A. craccivora* on plants' receiving various amounts of N fertilization, aphids'  $r_m$  was only affected by its fecundity rate. In other words, greater aphid fecundity by increasing N application resulted in higher  $r_m$  of aphids. Namely, according to Jauset *et al.* (2000), lower egg mortality, larger nymphs and females, and higher oviposition frequencies of *T. vaporariorum* result from over-fertilization of the host plant. The  $r_m$  of the whiteflies that developed on tomato plants fertilized with the highest nitrogen level was about 14% higher (no significance was found) than that of whiteflies that developed on plants supplied with the lowest nitrogen level. Besides, Khan and Port (2008) reported that while higher N fertilization levels did not significantly affect maturation time of *Rhopalosiphum padi* (L.) on *Triticum aestivum* L., aphids'  $r_m$  was increased in aphid fecundity on plants receiving higher N fertilization levels. Therefore, it seems that fecundity of insect herbivore is the most important determinant of population growth in the context of fertilizer effects (Stafford *et al.*, 2012).

Population size of *A. craccivora* responded well to N fertilization levels on amaranth plants. Chow *et al.* (2009) similarly reported that the density of *Tetranychus urticae* Koch on the cut roses was affected by N fertilization and enhanced with increasing N level (from 15 to 150 ppm). High population density of *B. tabaci* adult and immature instars on tomato were related to fertilization by 205 mg L<sup>-1</sup> N and 335 mg L<sup>-1</sup> (Žanić *et al.*, 2011). Moreover, the population growth of *A. gossypii* was at its highest on cucumber at the greatest N level (190 ppm) (Hosseini *et al.*, 2010a).

In general, the better the growth rate is, the higher the population density of *A. craccivora* is correlated with the highest N content, in plants fertilized with 100% of the recommended N level. Dietary N, which is an important factor in aphid development and growth, fundamentally exists in the forms of

amino acids in phloem (Douglas, 2006). Therefore, increasing N in plant tissues with N fertilization may enhance total amino acid concentration in phloem, thus, aphids have a greater supply of nutritional elements (White, 1993; Bentz *et al.*, 1995). On the other hand, increasing N fertilization decreased the C/N ratio of amaranth plants phloem tissue, and probably their carbon-based secondary compounds, including flavonoid (Ferrerres *et al.*, 2011), saponin and coumarine (Rufino Arcanjo *et al.*, 2011), and sterol glucoside compounds (Dinda *et al.*, 2006). Therefore, reduced levels of defensive metabolites of *G. globosa* plants induced by higher N content might have contributed to the better performance and population increase of *A. craccivora*. Yet, concentrations of carbon-based secondary metabolites of amaranth plants under different N regimes were not evaluated in the current study. Therefore, the possibility of this hypothesis would be addressed in future research.

The plant phenotypic plasticity is the ability of morphological modification in response to the biotic and abiotic stimulants (Valladares *et al.*, 2007; Žanić *et al.*, 2013). The biotic effect of aphids infestation on host plant and the abiotic response to high N fertilization led to changes in growth parameters of plants (Valladares *et al.*, 2007). In this study, vegetative (shoot to root ratio), and reproductive (flower number) yield, as well as physiological status of amaranth plants were greatly affected by N fertilization levels and aphid presence.

In the presence of aphid, the associated productivity losses of *G. globosa* plants which received the highest N fertilization was more than those of plants which were fed with lower N fertilization levels. For instance, the number of flowers in the plants with 100% of the recommended N fertilization showed greater loss (c.a 22%) than that in the plants receiving 60% N (c.a 5%). In addition, the higher loss of shoot to root ratio (c.a 59%) was acquired in the plants grown on 100% N level than that in plants grown with 60% N level (18%). The highest productivity losses of infested plants fertilized with 100 % N may be attributed to



their lowest net photosynthesis rate ( $P_n$ ) (Figure 1). Indeed, higher aphid population at higher N fertilization level could decrease  $P_n$  by feeding on sap phloem N content in which the lower amount of N element was allocated to plants  $P_n$  (Davies *et al.*, 2004). Similar to our result, Hosseini *et al.* (2010a) reported that at the highest N fertilization level (190 ppm), *A. gossypii* induced the greatest yield reduction of cucumber plants, which is probably related to a significant decrease in net photosynthesis rate.

*G. globosa* is an ornamental plant used as a bed in public locations or cut flower; therefore, it is critical to be grown in a way that supports lower risks of insecticide applications. In floriculture crops, reduction of fertilization could be considered as an effective tactic in pest management, if altered fertilization regimes can reduce pest populations with little loss in crop yield (Chow *et al.*, 2009; 2011). Our results have demonstrated that lowering N fertilization in amaranth plants with 60% of recommended N significantly decreased *A. craccivora* population without substantial loss of flower yield and marketability. Then, a proper application of N fertilization could be used for reducing *A. craccivora* populations. Consequently, growers would benefit from application of less insecticides. Moreover, reduction in the amount of N application has additional advantages such as reducing floriculture production costs and chemical run-off. Finally, we suggest that future studies should evaluate the effectiveness of reduction in fertilization as a relatively simple and easily implemented tactic with other IPM strategies, including biological control (Hosseini *et al.*, 2010b), and cultural practices (Stavisky *et al.*, 2002) for *A. craccivora* management on amaranth plants and other ornamental crops.

#### ACKNOWLEDGEMENTS

The authors greatly acknowledge Parisa Taheri from Ferdowsi University of Mashhad for helpful comments on the manuscript. This research was supported by Ferdowsi University of Mashhad.

#### REFERENCES

1. Alfonsina, S. 2008. Aphididae (Hemiptera) on Ornamental Plants in Cordoba (Argentina). *Rev. Soc. Entomol. Argent.*, **67**: 49–56.
2. Aqueel, M. A. and Leather, S. R. 2011. Effect of Nitrogen Fertilizer on the Growth and Survival of *Rhopalosiphum padi* (L.) and *Sitobion avenae* (F.) (Hom.: Aphididae) on Different Wheat Cultivars. *Crop Prot.*, **30**: 216–221
3. Bachman, G. 2011, April 11. Tough Gomphrena Makes Good Addition to Gardens. Retrieved April 25, 2013.
4. Benary, E. [Internet]. 2011. *Gomphrena globosa*. Benary, Retrieved from April 15 2013 from: www.benary.com
5. Bentz, J. and Larew, H. 1992. Ovipositional Preference and Nymphal Performance of *Trialeurodes vaporariorum* (Homoptera: Aleyrodidae) on *Dendranthema grandiflora* (Tzvelev) Under Different Fertilizer Regimes. *J. Econ. Entomol.*, **85**: 514–517.
6. Bentz, J. A., Reeves, J., Barbosa, P. and Francis, B. 1995. The Effect of Nitrogen Fertilizer Effect on Selection, Acceptance, and Suitability of *Euphorbia pulcherrima* (Euphorbiaceae) as a Host plant to *Bemisia tabaci* (Homoptera: Aleyrodidae). *Environ. Entomol.*, **24**: 40–45.
7. Birch, L. 1948. Intrinsic Rate of Natural Increase of an Insect Population. *J. Anim. Ecol.*, **17**: 15–26.
8. Byrne, D. N., Cohen, A. C. and Draeger, E. A. 1990. Water Uptake From Plant Tissue By the Egg Pedicel of the Greenhouse Whitefly *Trialeurodes vaporariorum* (Westwood) (Homoptera: Aleyrodidae). *Canad. J. Zool.*, **68**: 1193–1195.
9. Carey, J. R. 1993. Applied Demography for Biologists with Special Emphasis on Insects. Oxford University Press, New York, 206 PP.
10. Chau, A., Heinz, K. M. and Davies, F. T. 2005. Influences of Fertilization on Population Abundance, Distribution and Control of *Frankliniella occidentalis* on Chrysanthemum. *Entomol. Exp. Appl.*, **117**: 27–39.
11. Chow, A., Chau, A. and Heinz, K. M. 2009. Reducing Fertilization For Cut Roses: Effect on Crop Productivity and Twospotted Spider Mite Abundance, Distribution, and



- Management. *Hort. Entomol.*, **102**: 1896 – 1907.
12. Chow, A., Chau, A. and Heinz, K. M. 2011. Reducing Fertilization: A Management Tactic Against Western Flower Thrips on Roses. *J. Appl. Entomol.*, **136**: 506–529.
  13. Davies, F. T. J., He, C.J., Chau, A., Heinz, K. M. and Cartmill, A. D. 2004. Fertility Affects Susceptibility of Chrysanthemum to Cotton Aphid (*Aphis gossypii*), Influence on Plant Growth, Photosynthesis, Ethylene Evolution and Herbivore Abundance. *J. Am. Soc. Hort. Sci.*, **129**: 344–353.
  14. Dinda, B., Ghosh, B., Achari, B., Arima, S., Sato, N. and Harigaya, Y. 2006. Chemical Constituents of *Gomphrena globosa*. *Nat. Pro. Sci.*, **12**: 89–93.
  15. Douglas, A. E. 2006. Phloem–sap Feeding by Animals: Problems and Solutions. *J. Exp. Bot.*, **57**: 747–754.
  16. Elliott, M. L., Broschat, T. K., Uchida, J. Y. and Simone, G. W. 2004. *Compendium of Ornamental Palm Diseases and Disorders*. APS Press, St. Paul, MN, 69 PP.
  17. Elser, J. J., Fagan, W. F., Denno, R. F., Dobberfuhl, D. R., Folarin, A., Huberty, A., Interlandi, S., Kilham, S. S., McCauley, E., Schulz, K. L., Siemann, E. H. and Sterner, R. W. 2000. Nutritional Constraints in Terrestrial and Freshwater Food Webs. *Nature*, **408**: 578–580.
  18. Ferreres, F., Gil-Izquierdo, A., Valentão, P. and Andrade, B. P. 2011. Structural Characterization of Phenolics and Betacyanins in *Gomphrena globosa* by High-Performance Liquid Chromatographydiode Array Detection/Electrospray Ionization Multi-Stage Mass Spectrometry. *Rap. Community Mass. Spectrom.*, **25**: 3441–3446.
  19. Fallahpour, F., Ghorbani, R., Nassiri Mahallati, M. and Hosseini, M. 2015. Demographic Parameters of Lipaphis Erysimion Canola Cultivars under Different Nitrogen Fertilization Regimes. *J. Agric. Sci. Tech.* **17**:35-47.
  20. Ghasemi, Gh. M. and Kafi, M. 2007. *Scientific and Practical Floriculture*. Esfahan Golbon Press. (in Persian)
  21. Hogendorp, B., Cloyd, R. and Swiader, J. 2006. Effect of Nitrogen Fertility on Reproduction and Development of Citrus Mealybug, *Planococcus citri* Risso (Homoptera: Pseudococcidae), Feeding on Two Colors of Coleus, *Solenostemon scutellarioides* L. *Codd. Environ. Entomol.*, **35**: 201–211.
  22. Hosseini, M., Ashouri, A., Enkegaard, A., Goldansaz, S. H., Nassiri Mahalati, M. and Hosseinaveh, V. 2010a. Performance and Population Growth Rate of the Cotton Aphid and Associated Yield Losses of Cucumber Under Different Nitrogen Fertilization Regimes. *Int. J. Pest. Manag.*, **56**: 127–137.
  23. Hosseini, M., Ashouri, A., Enkegaard, A., Wolfgang, W. W., Goldansaz, S. H., Nassiri Mahalati, M. and Sarraf Moayeri, H. R. 2010b. Plant Quality Effects on Intraguild Predation Between *Orius laevigatus* Fieber (Hemiptera: Anthocoridae) and *Aphidoletes aphidimyza* (Rondani) (Diptera: Cecidomyiidae). *Entomol. Exp. Appl.*, **135**: 208 – 216.
  24. Huang, J. S., Nelson, P. V., Bailey, D. A., Fonteno, W. C. and Mingis, N. C. 2002. Assessment of the Need for Nitrogen, Phosphorus, Potassium, and Sulfur Preplant Nutrients for Plug Seedling Growth. *Hort. Sci.*, **37**: 529–533.
  25. Jauset, A. M., Sarasua, M. J., Avilla, R. and Albajes, R. 2000. Effect of Nitrogen Level Applied to Tomato on the Greenhouse Whitefly. *Crop Prot.*, **19**: 255–261.
  26. Jones, C. E. 1984. AOAC Official Analytic Chemists. In: " *Animal Feed* ", (Eds.): Wiliams, S.. Arlington, VA, PP.155–156.
  27. Jones, M. G. 1967. Observation on Two Races of the Groundnut Aphid, *Aphis craccivora*. *Entomol. Exp. Appl.*, **10**: 31–38.
  28. Khan, M. and Port, G. 2008. Performance of Clones and Morphs of Two Cereal Aphids on Wheat Plants With High and Low Nitrogen Content. *Entomol. Sci.*, **11**: 159- 165.
  29. Lovisolo, O. and Conti, M. 1966. Identification of an Aphid–transmitted *Cowpea Mosaic Virus*. *Neth. J. Plant Path.*, **72**: 265–269.
  30. Maia, A. H. N., Luiz, A. J. B. and Campanhola, C. 2000. Statistical Inference on Associated Fertility Life Parameters Using Jackknife Technique: Computational Aspects. *J. Econ. Entomol.*, **93**: 511–518.
  31. Mattson, W. J. J. 1980. Herbivory in Relation to Plant Nitrogen Content. *Ann. Rev. Ecol. Syst.* **11**: 119–161.
  32. Mc Neill, S. and Southwood, T. R. E. 1978. Aspects of Plant and Animal Coevolution. In: "*The Role of Nitrogen in the Development of Insect/Plant Relationships*", (Eds.): Harborne, J. S.. Academic Press, London, PP. 77– 98.

33. Mengel, K. and Kirkby, E. A. 2001. *Principles of Plant Nutrition*. 5<sup>th</sup> Edition, Kluwer Academic Publishers, Boston, USA.
34. Meyer, J. S., Ingersoll, C. G., McDonald, L. L. and Boyce, M. S. 1986. Estimating Uncertainty in Population Growth Rates: Jackknife vs. Bootstrap Techniques. *Ecol.*, **67**: 1156–1166.
35. Minkenbergh, O. P. J. M. and Fredrix, M. J. J. 1989. Preference and Performance of an Herbivorous Fly, *Liriomyza trifolii* (Diptera: Agromyzidae), on Tomato Plants Differing in Leaf Nitrogen. *Ann. Entomol. Soc. America*, **82**: 350–354.
36. Obopile, M. 2006. Economic Threshold and Injury Levels for Control of Cowpea Aphid, *Aphis craccivora* L. (Homoptera: Aphididae) on Cowpea. *Afr. Plant Prot.*, **12**: 111–115.
37. Ponder, K. L., Pritchard, J., Harrington, R. and Bale, J. S. 2000. Difficulties in Location and Acceptance of Phloem Sap Combined With Reduced Concentration of Phloem Amino Acids Explain Lowered Performance of the Aphid *Rhopalosiphum padi* on Nitrogen Deficient Barley (*Hordeum vulgare*) Seedlings. *Entomol. Exp. Appl.*, **97**: 203–210.
38. Richards, L. A. 1954. Diagnosis and Improvement of Saline and Alkali Soils. Handbook No. 60, USDA.
39. Rufino Arcanjo, D. D., Albuquerque, A. C., Neto, M. B., Resende Santana, L. C., Branco Silva, N. C., Moita, M. M. and Medeiros, M. 2011. Phytochemical Screening and Evaluation of Cytotoxic, Antimicrobial and Cardiovascular Effects of *Gomphrena globosa* L. (Amaranthaceae). *J. Med. Plants Res.*, **5**: 2006–2010.
40. SAS Institute. 2003. SAS users guide: statistics. SAS Institute, Cary, NC.
41. Stafford, D. B., Tariq, M., Wright, D. J., Rossiter, J. T., Kazana, E., Leather, S. R., Ali, M. and Staley, J. T. 2012. Opposing Effects of Organic and Conventional Fertilizers on the Performance of a Generalist and a Specialist Aphid Species. *Agric. For. Entomol.*, **14**: 270–275.
42. Stavisky, J., Funderburk, J., Brodbeck, B. V. and Olson, S. M. 2002. Population Dynamics of *Frankliniella* spp. and Tomato Spotted Wilt Incidence as Influenced by Cultural Management Tactics in Tomato. *J. Econ. Entomol.*, **95**: 1216–1221.
43. Tao, C. C. 1990. Aphid–Fauna of Taiwan Province, China: Taiwan Provincial Museum, Taipei.
44. Trdan, S., Milevoj, L., Žežline, I., Raspudič, E., Andus, L., Vidrih, M., Bergant, K., Valič, N. and Žnidarčič, D. 2005. Feeding Damage by Onion Thrips, *Thrips tabaci* Lindeman (Thysanoptera, Thripidae), on Early White Cabbage Grown under Insecticide-Free Conditions. *Afr. Entomol.*, **13(1)**: 85–95.
45. Trdan, S., Žnidarčič, D., Kač, M. and Vidrih, M. 2008. Yield of Early White Cabbage Grown under Mulch and Non-Mulch Conditions with Low Populations of Onion Thrips (*Thrips tabaci* Lindeman). *Int. J. Pest. Manag.*, **54(4)**: 309–318.
46. Valladares, F., Gianoli, E. and Gomez, J. 2007. Ecological Limits to Plant Phenotypic plasticity. *New Phytol.*, **176**: 749–763.
47. Vehrs, S. L., Walker, G. P. and Parrella, M. P. 1992. Comparison of Population Growth Rate and Within–Plant Distribution Between *Aphis gossypii* and *Myzus persicae* (Homoptera: Aphididae) Reared on Potted Chrysanthemums. *J. Econ. Entomol.*, **85**: 799–807.
48. White, T. C. R. 1993. The Inadequate Environment: Nitrogen and the Abundance of Animals. New York, Springer Verlag, Berlin.
49. Žanić, K., Ban, D., Gotlin Čuljak, T., Goreta Ban, S., Dumičić, G., Haramija, J. and Žnidarčič, D., 2013. Aphid Populations (Hemiptera:Aphidoidea) Depend of Mulching in Watermelon Production in the Mediterranean Region of Croatia. *Span. J. Agric. Res.*, **11(4)**: 1120–1128.
50. Žanić, K., Dumičić, G., Škaljac, M., Goreta Ban, S. and Urlič, B. 2011. The Effects of Nitrogen Rate and the Ratio of  $\text{NO}_3^-:\text{NH}_4^+$  on *Bemisia tabaci* Populations in Hydroponic Tomato Crops. *Crop. Protect.*, **30**: 228–233.
51. Zehnder, C. B. and Hunter, M. D. 2008. Effect of Nitrogen Deposition on the Interaction Between an Aphid and Its Host Plant. *Ecol. Entomol.*, **33**: 24–30.



## اثر کوددهی نیتروژن بر ویژگی های زیستی شته *Aphis craccivora* (Hemiptera: Aphidiae) و کاهش عملکرد گل تکمه ای

۱. حسینی، م. حسینی، م. گلدانی، ج. کریمی، و ح. مددی

### چکیده

از روش های مؤثر در مدیریت آفات، افت کیفیت گیاه میزبان از طریق کاهش سطح تغذیهی نیتروژن می- باشد. در این پژوهش، توانایی زیستی و نرخ رشد جمعیت شته *Aphis craccivora* (Hemiptera: Aphidiae) تحت مقادیر مختلف کود نیتروژن و اثر برهمکنش کوددهی و فراوانی شته بر عملکرد و ویژگی های بازارپسندی گیاه زینتی گل تکمه ای *Gomphrena globosa* L. در شرایط گلخانه ای بررسی گردید. چهار تیمار کود نیتروژن شامل صفر، ۳۰، ۶۰ و ۱۰۰ درصد سطح توصیه شدهی کوددهی تعیین شد. نتایج نشان داد، نرخ ذاتی افزایش جمعیت شته در بالاترین سطح کوددهی (۱۰۰ درصد سطح توصیه شده) به طوری معنی- داری افزایش یافت. هم چنین فراوانی و نرخ رشد جمعیت شته با کوددهی نیتروژن همبستگی مثبت نشان داد. برهمکنش حضور شته و کوددهی بر عملکرد گیاه میزبان تأثیری معنی دار داشت. در شرایط عدم حضور شته، عملکرد گیاه با افزایش سطح کوددهی نیتروژن به طوری معنی داری افزایش یافت. با این وجود، در بالاترین سطح توصیه شده کوددهی (۱۰۰ درصد) جمعیت شته، تعداد گل ها و نسبت زیست توده اندام هوایی به ریشه را به شدت کاهش داد. براساس نتایج مطالعهی حاضر، اعمال کوددهی بهینه به عنوان تاکتیک مدیریتی کارآمد در کنترل انبوهی جمعیت شته افاقیا روی گیاه زینتی گل تکمه ای توصیه می گردد.