

***Aphis nerii* and its parasitoid *Aphidius colemani* on oleander as a model for assessing lead pollution effects**

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Abstract

Oleander *Nerium oleander* L. has generally been used as biomonitor plant for atmospheric lead (Pb) pollution. Therefore, it was exploited to investigate the effects of Pb pollution to the oleander aphid *Aphis nerii* Boyer de Fonscolombe and its associated parasitoid *Aphidius colemani* Viereck. Pb concentrations in plant leaves collected from areas of low (natural reserve), intermediate (public garden) and high (gasoline station near crowded road) levels of pollution were (4.0, 7.1 and 9.2 ppm), respectively, while it was 1.3 ppm in the control. Some biological variables of *A. nerii* and its parasitoid, *A. colemani* raised on plant leaves representing the three levels of Pb pollution were investigated. In most variables studied, Pb-treatments were differing from the control for both insects. Development times and mortality percentages of both insects were positively correlated with the Pb concentration and with the Pb exposure frequency as well. On the contrary, fecundity and longevity were negatively correlated with the Pb concentration and with the Pb exposure frequency. In conclusion, the Pb would strongly affect insects' biology, and this effect was of a residual pattern; it affected the pest insect and subsequently its parasitoid. Moreover, Pb has accumulative and chronic effects; since it was more conspicuous when insects frequently exposed to Pb rather than when they were exposed once to Pb.

Key words: *Aphidius colemani*, *Aphis nerii*, environmental pollution, lead, *Nerium oleander*.

Introduction

Lead (Pb) is one of the most common and the most toxic heavy metals in the environment (Xia, 2004). Living systems of at least 100 countries are still exposed to considerable Pb concentrations owing to air pollution despite the banning of Pb in gasoline (Kaya *et al.*, 2010). It has no biological functions in organisms including plants (Li *et al.*, 2007); on the contrary, it has an adverse impact on plants, animals and humans even when absorbed in small amounts (Li *et al.*, 2007; Azmat *et al.*, 2009; Rai *et al.*, 2010). Heavy metals, including Pb, can also affect plant quality (Lill *et al.*, 2002); it inhibits chlorophyll biosynthesis (Xiong, 1997a). Plants can accumulate Pb via root and shoot, and the concentrations in plant tissues are significantly related to the Pb levels in the environment (Xiong, 1997b). Variation in host-plant quality influences insect herbivore survival (Ladner and Altizer, 2005), development time (Wheeler, 2001) and fecundity (Tsai and Wang, 2001). So, interest in determination of such effects of heavy metals on biological systems is becoming of vital importance (Mojica *et al.*, 2005) in order to increase awareness about its harmful effects on living organisms. The animals most commonly used as biomonitors for pollution are bivalve mollusks (Elder and Collins, 1991); the use of such mollusks is limited for many constraints (Clarke, 1981; Pennak, 1989; Couillard *et al.*, 1993). Insects have been proposed as an alternative (Hare and Tessier, 1996), since insects seem to display the greatest diversity of biochemical and genetic responses with stress from exposure to exogenous chemicals; therefore, they are considered a valid model for quantifying the impacts of xenobiotics (Landa *et al.*, 1991; Hare, 1992). Among insects, aphids are ideal organisms to study such effects (Zehnder and Hunter, 2007), because of the lack of ge-

netic variation across individuals and generations, in which the susceptibility of the individuals to the same Pb concentration is almost the same.

Oleander (*Nerium oleander* L.) is an evergreen shrub that is widely cultivated as an ornamental in warm areas of the world (Blackman and Eastop, 1984). It is commonly used for biomonitoring of atmospheric pollution (Sawidis *et al.*, 2001; Oliva and Mingorance, 2004; Hemdez *et al.*, 2005) for its ability to absorb and store heavy metals, particularly Pb (Kaya *et al.*, 2010; Shehu *et al.*, 2010). In the countries of origin, oleander is attacked by many pests: scale insects, mealybugs and aphids, *Aphis nerii* Boyer de Fonscolombe (Rothschild, 1972). The aphid is the most important one; it sucks saps from leaves and fruits, the damage are mainly aesthetic due to the honeydew and the resulting black sooty mold. *A. nerii* has been implicated in the transmission of at least 4 plant viruses (Chan *et al.*, 1991). *A. nerii* reproduce exclusively by parthenogenesis; they are all females, and produce clones of themselves by self fertilization (Blackman and Eastop, 1984; Harrison and Mondor, 2011). One of the parasitoids associated with this aphid species is *Aphidius colemani* Viereck (Sary, 1988). The use of bioindicators has proved to be effective to detect heavy metals impacts on organisms (Divan-Junior *et al.*, 2009). Therefore, the tritrophic interaction among the oleander plant and its associated aphid pest and the parasitoid was used to investigate the effects of Pb on the insects.

Materials and methods

Pb concentrations determination

Leaves of oleander were randomly detached from plants grown in different locations with expected pollu-

tion with different Pb concentrations. However, the low Pb pollution level expected in the natural reserves (32°5'14.19"N 35°39'40.93"E), intermediate Pb pollution level expected in public gardens that far from both crowded roads and industrial areas (32°2'47.39"N 35°46'43.65"E), and high Pb pollution level expected in gasoline stations located at crowded roads nearest to industrial areas (32°0'56.29"N 35°50'40.35"E). Plants grown on the campus greenhouses (32°1'26.43"N 35°42'57.93"E) having almost zero Pb concentrations, served as a control treatment. The leaf samples; 0.5 Kg each, representing the control and the 3 Pb levels (in triplicate) were transferred in plastic bags to the laboratory. Leaves were washed well with distilled water, dried at 75 °C for 5 days and ground with an electrical mill (Moulinex-3, France). The samples were digested with concentrated HNO₃-HClO₄ and then analyzed following the method used by Kaya *et al.* (2010). Moreover, Pb concentrations were determined by dry-ashing using atomic-absorption spectrophotometer (Shimadzu AA-680, Japan).

Colonies of aphids and parasitoids

Four colonies of aphid were initiated from the greenhouse aphid colony; one was raised on non-polluted oleander plants held on the campus greenhouses (aphid control colony). The aphids for the other 3 colonies were raised for many generations on plants left in the 3 previously mentioned locations to be polluted with the different Pb concentrations (aphids frequently exposed to Pb colonies). For the treatments of the aphids exposed once to Pb, insects were collected from the aphid control colony and were fed once on oleander leaves detached from the 3 Pb-polluted locations. Seven cultures of the parasitoid were initiated and raised on the aforementioned aphid colonies.

Conditions of experimenting

Both aphids and parasitoids used in the experiments were maintained at 20 ± 2 °C; which is the optimal temperature for both the aphid and the parasitoid as reported by Ozder and Saglam (2013) and van Steenis (1993), respectively. The relative humidity was 65 ± 5%, and the light regime was 16L:8D. In addition, such conditions were chosen to conduct our experiments to enable us to compare our control results with the results obtained by Ozder and Saglam (2013) (for the aphid) and van Steenis (1993) (for the parasitoid).

Treatments on aphids

Aphids were collected from the aforementioned established colonies and fed on the Pb-free or the Pb-polluted leaves (of the 3 Pb concentrations) according to the corresponding treatment. Nymph development time, female fecundity and longevity, mortality, weights of wingless adult offspring and production of alate individuals were estimated following the methodology used by Ozder and Saglam (2013). Females were weighed using AE ADAMS®-UK balance (max = 210 g, d = 0.0001 g). Average weight of 10 females for each treatment was considered, while 4 individual aphids were used as 4 replicates for investigating the other variables.

Calculation of the intrinsic rate of increase

Intrinsic rate of increase (r) was calculated using data of 3 replications for 5 generations. Apterous adult of *A. nerii* were picked up by camel hair brush from the source plant (according to the treatment to be tested: control, 3 Pb conc., exposed once or exposed frequently) and were settled on the experimental leaves. The newly born nymphs were considered the first generation, and then they were released into new leaves to have the second generation, and so on until reached the fifth generation. The duration between nymph birth and its first laid progeny (d) and number of progeny (M_d) per female were recorded. The intrinsic rate of increase was calculated according to the equation: $r = c (\log_e M_d)/d$, where c : correcting constant = 0.74 (Wyatt and White, 1977).

Treatments on the parasitoids

Parasitoids for the initial culture were aspirated from the emerged *A. nerii* from mummies collected from the greenhouse oleanders. Parasitoids of the experiments were investigated using the aforementioned groups of aphids. The development period, sex ratio, and mortality of the parasitoid were estimated following the methodology used by van Steenis (1993). As for the female parasitoid, fecundity and longevity were also investigated by 4 replicates. Intrinsic rate of parasitoid increase was calculated using data of 3 generations.

Statistical analysis

The statistical analysis was performed using the statistics software JMP version 8. Differences in both aphid and parasitoid biology at different Pb concentrations and different exposure frequency were tested by the analyzing of the variance (Wilkinson, 1990). When significant differences were detected at different Pb concentrations, the means were compared to each other using Tukey HSD at 0.05 probability level (Zar, 1999). The Student T-test was used to compare two means of insects exposures (exposure once and exposed many times). Finally, the correlation analysis was performed using the proc GLM of the statistical package SigmaStat version 16 (SPSS, 1997).

Results

The expected low Pb pollution level was found to be 4.0 ppm Pb concentration, the intermediate level was 7.1 ppm, and the high level was 9.2 ppm. The Pb concentration in the control treatment was 1.3 ppm.

The aphid biology is obviously affected by the Pb-pollution. In most variables studied, Pb-treatments were different from the control treatments. The highest Pb concentration in the plant leaves prolonged the aphid development period from birth to adulthood (table 1). Aphid nymphs frequently fed on leaves polluted with Pb took extra 1-1.5 days to reach adult stage than nymphs exposed only once to Pb at all concentrations tested ($t(1) = 2.07$, $P < 0.0001$) (table 1). Moreover, aphid fecundity was reduced due to the increasing Pb concentration ($F_{3,8} = 16.0$, $P < 0.0001$), whereas there were no

Table 1. *A. nerii* development, fecundity, longevity, mortality, weights of offspring and alate production in relation to different lead (Pb)-pollution concentrations under two patterns of Pb-exposure frequency using oleander plants. Number of replicates = 4.

Variable	Exposure frequency to Pb	1.3 ppm Pb	4.0 ppm Pb	7.1 ppm Pb	9.2 ppm Pb
		(Control) Mean ± SE	(Low) Mean ± SE	(Intermediate) Mean ± SE	(High) Mean ± SE
Nymph development (days)	Exposed once	12.3 ± 0.18 b	12.7 ± 0.18 b	12.8 ± 0.18 b	13.8 ± 0.18 a
	Previously exposed	12.3 ± 0.15 c	13.8 ± 0.15 b	14.3 ± 0.15 b	15.3 ± 0.15 a
Fecundity/♀	Exposed once	16.0 ± 0.2 a	15.0 ± 0.2 b	15.0 ± 0.2 b	14.0 ± 0.2 c
	Previously exposed	16.0 ± 0.15 a	14.0 ± 0.15 b	13.0 ± 0.15 c	13.0 ± 0.15 c
♀ longevity (days)	Exposed once	29.6 ± 0.15 a	29.1 ± 0.15 ab	28.8 ± 0.15 b	28.8 ± 0.15 b
	Previously exposed	29.6 ± 0.15 a	28.3 ± 0.15 b	28.0 ± 0.15 b	27.7 ± 0.15 b
Mortality (nymphs & ♀♀)	Exposed once	23% ± 0.16 c	24% ± 0.16 b	24% ± 0.16 b	25% ± 0.16 a
	Previously exposed	23% ± 0.15 c	24% ± 0.15 b	27% ± 0.15 a	27% ± 0.15 a
Weights of adult offspring (mg)	Exposed once	0.65 ± 0.02 a	0.60 ± 0.02 ab	0.57 ± 0.02 b	0.55 ± 0.02 b
	Previously exposed	0.65 ± 0.02 a	0.56 ± 0.02 b	0.53 ± 0.02 b	0.52 ± 0.02 b
Alate production	Exposed once	0.0% b	0.0% b	0.0% b	2.0% ± 0.07 a
	Previously exposed	0.0% d	1.0% ± 0.13 c	5.0% ± 0.13 b	12% ± 0.13 a

Means with different letters within the same row are significantly different at 5% error level.

Table 2. Aphid parasitoid *A. colemani* development, fecundity, longevity, mortality and sex ratio raised on aphids fed on oleander plants exposed to different lead (Pb)-pollution levels under two patterns of Pb-exposure frequency. Number of replicates = 4.

Variable	Aphid (parasitoid host) exposure frequency to Pb	Aphids fed on oleander plants			
		1.3 ppm Pb (Control) Mean ± SE	4.0 ppm Pb (Low) Mean ± SE	7.1 ppm Pb (Intermediate) Mean ± SE	9.2 ppm Pb (High) Mean ± SE
Development (days)	Exposed once	12.1 ± 0.17 c	13.15 ± 0.17 b	13.6 ± 0.17 b	14.6 ± 0.17 a
	Previously exposed	12.1 ± 0.13 d	15.0 ± 0.13 c	15.5 ± 0.13 b	17.4 ± 0.13 a
Fecundity/♀	Exposed once	281 ± 3.3 a	260 ± 3.3 b	251 ± 3.3 bc	243 ± 3.3 c
	Previously exposed	281 ± 3.1 a	242 ± 3.1 b	233 ± 3.1 b	210 ± 3.1 c
♀ longevity (days)	Exposed once	5.5 ± 0.18 a	5.6 ± 0.18 a	5.3 ± 0.18 a	5.3 ± 0.18 a
	Previously exposed	5.5 ± 0.17 a	5.2 ± 0.17 a	5.2 ± 0.17 a	5.0 ± 0.17 a
Mortality	Exposed once	17% ± 0.7 b	16% ± 0.7 b	18% ± 0.7 ab	20.8% ± 0.7 a
	Previously exposed	17% ± 1.3 a	20% ± 1.3 a	22% ± 1.3 a	22% ± 1.3 a
Offspring sex ratio	Exposed once	0.57 ± 0.01 b	0.59 ± 0.01 ab	0.63 ± 0.01 ab	0.64 ± 0.01 a
	Previously exposed	0.57 ± 0.01 c	0.69 ± 0.01 b	0.72 ± 0.01 b	0.78 ± 0.01 a

Means with different letters within the same row are significantly different at 5% error level.

significant differences in female longevity among the different Pb concentrations except for the control ($F_{3,8} = 6.22, P < 0.0086$). High mortalities were resulted at the highest Pb concentration for both patterns of aphid exposure, and even the low concentration of Pb caused significant mortalities compared with the control ($F_{3,8} = 27.6, P < 0.0001$). The weight of aphid offspring was not related to Pb concentration for both exposure patterns but they were different from the control ($F_{3,8} = 15.9, P < 0.0002$). When aphid exposed once to Pb, an increase in the production of alate individuals was recorded at the highest Pb concentration, while the two other Pb concentrations were at bar with the control ($F_{3,8} = 218.2, P < 0.0001$). Moreover, an increase in alate production was noticed as a result of the increase in the Pb concentration when mothers were frequently exposed to Pb ($F_{3,8} = 1780, P < 0.0001$) (table 1). Accumulative effect of Pb on the intrinsic rate of aphid increase was found; the rate decreased with increasing the exposure

frequency of the Pb ($t(2) = 421.13, P < 0.0001$). For all variables investigated, aphids were affected when frequently exposed to Pb as compared with those exposed only once to Pb (table 3). In addition, the parasitoid was remarkably affected by the Pb-pollution except for the longevity and mortality; Pb-treatments were different from the control treatments. Parasitoid development period was affected by the Pb-pollution level compared with the control. A longer development period was obtained at the higher Pb concentration ($F_{3,8} = 281.5, P < 0.0001$). The development periods at low and intermediate Pb concentrations were at bar with each other when parasitoids fed on aphids exposed once to Pb ($F_{3,8} = 37.1, P < 0.0001$) (table 2). Parasitoid fecundity at the highest Pb concentration was lower than those at both low and intermediate Pb concentrations and all differed from the control ($F_{3,8} = 92.9, P < 0.0001$). Unlike all the variables tested, longevity of parasitoid was not affected neither by the Pb pollution concentrations nor

Table 3. Effect of Pb-exposure frequency on development, fecundity, longevity, mortality, intrinsic rate of increase and other variables of the aphid *A. nerii* and its parasitoid *A. colemani* using oleander plant.

Variable	Exposure frequency to Pb	For aphid	For parasitoid
Development (days) ⁽¹⁾	Exposed once	13.1 ± 0.19 b	13.8 ± 0.27 b
	Previously exposed	14.5 ± 0.19 a	15.9 ± 0.27 a
Fecundity/♀ ⁽¹⁾	Exposed once	14.7 ± 0.17 a	251.3 ± 3.7 a
	Previously exposed	13.3 ± 0.17 b	228.3 ± 3.7 b
Longevity (days) ⁽¹⁾	Exposed once	28.9 ± 0.1 a	5.400 ± 0.1 a
	Previously exposed	28.0 ± 0.1 b	5.13 ± 0.1 a
Mortality ⁽¹⁾	Exposed once	24.3% ± 0.33 b	18.2% ± 0.7 b
	Previously exposed	26.0% ± 0.33 a	21.3% ± 0.7 a
Weights of adult offspring (mg) ⁽¹⁾	Exposed once	0.57 ± 0.01 a	-
	Previously exposed	0.54 ± 0.01 b	-
Alate production ⁽¹⁾	Exposed once	0.67 ± 1.0 b	-
	Previously exposed	6.00 ± 1.0 a	-
Offspring sex ratio ⁽¹⁾	Exposed once	-	0.62 ± 0.01 b
	Previously exposed	-	0.73 ± 0.01 a
Intrinsic rate of increase, r (females per female per day) ⁽²⁾	Exposed once	0.293 ± 0.003 b	0.28 ± 0.01 a
	Previously exposed	0.235 ± 0.003 c	0.18 ± 0.01 b
	Control	0.361 ± 0.003 a	0.32 ± 0.01 a

⁽¹⁾ Means with different letters for the same insect species within the same variable are significantly different at 5% error level using T-test (effect of Pb-exposure frequency)

⁽²⁾ Means with different letters for the same insect species in the same column are significantly different at 5% error level (effect of Pb-exposure frequency) using Tukey HSD

Table 4. Correlation analysis of the lead concentration and the exposure frequency to the lead versus the development, fecundity, longevity and mortality of *A. nerii* and the associated parasitoid, *A. colemani* using oleander plant.

Correlated variables	Insect	R value	Significance
Pb concentration vs. development	Aphid	0.678**	0.000
	Parasitoid	0.691**	0.000
Pb concentration vs. fecundity	Aphid	-0.667**	0.000
	Parasitoid	-0.717**	0.000
Pb concentration vs. longevity	Aphid	-0.572**	0.001
	Parasitoid	-0.383*	0.044
Pb concentration vs. mortality	Aphid	0.785**	0.000
	Parasitoid	0.618**	0.000
Exposure frequency vs. development	Aphid	0.817**	0.000
	Parasitoid	0.889**	0.000
Exposure frequency vs. fecundity	Aphid	-0.848**	0.000
	Parasitoid	-0.836**	0.000
Exposure frequency vs. longevity	Aphid	-0.870**	0.000
	Parasitoid	-0.402*	0.034
Exposure frequency vs. mortality	Aphid	0.689**	0.000
	Parasitoid	0.616**	0.000

*Significant correlation at 5% error level; **Significant correlation at 1% error level.

by both frequencies of exposure ($F_{3,8} = 1.57$, $P < 0.248$) (table 3). As for the parasitoid exposed once to Pb, higher mortality was recorded at the highest Pb concentration ($F_{3,8} = 7.81$, $P < 0.004$). Furthermore, mortality is statistically the same regardless of the Pb concentration when the parasitoid was frequently exposed to aphid fed on Pb ($F_{3,8} = 3.53$, $P < 0.0487$). Parasitoid offspring was shifted toward males due to the higher Pb pollution at both exposure patterns ($F_{3,8} = 45.0$, $P < 0.0001$) (table 2). The intrinsic rate of population increase, when the parasitoid was exposed frequently to Pb, was, by far, lower than those for both the control ($F_{3,8} = 47.4$, $P < 0.0001$)

and the parasitoid exposed only once (table 3). The correlation analysis of Pb concentration versus the biological variables of insects revealed that all the correlations were significant at the 1% error level for both insects. The parasitoid longevity (versus the Pb concentration and exposure frequency) was also significant at 5% error level ($r(26) = 0.40$, $P < 0.05$) as shown in (table 4). Moreover, the same trend of results was noticed for the correlation analysis of Pb-frequency of exposure versus the biological variables of both insects. The development period ($r(26) = 0.82$, $P < 0.01$) and mortality ($r(26) = 0.69$, $P < 0.01$) of both insects were positively

correlated with the Pb concentration and exposure frequency. On the other hand, fecundity ($r(26) = 0.85$, $P < 0.01$) and longevity ($r(26) = 0.87$, $P < 0.01$) were negatively correlated with the Pb concentration and the exposure frequency as shown in table 4.

Discussion

The lead (Pb) is a ubiquitous environmental toxicant which has been reported to have the highest concentration in organisms among most heavy metals (Lagrana *et al.*, 2011), its concentration among plants remains relatively unchanged with time (Allaway, 1968; David, 2003). It has growth-retarding effects (Cohn *et al.*, 1992); these effects were reflected on many insects in the different trophic levels (Warrington, 1987; Parke *et al.*, 1991; Watson, 1999; Haq *et al.*, 2011). Here we present the Pb effects using model insects of two levels: herbaceous (the aphid *A. nerii*) and entomophagous (the parasitoid *A. colemani*). The effects of the Pb concentration and the frequency of exposure to Pb were examined on the biology of an insect pest (aphid) and its parasitoid.

The highest concentration of the Pb in the host plant prolonged the nymph development and the frequent exposure to Pb maximized the differences of nymph development periods for the 3 Pb concentrations and the control. All the Pb concentrations decreased significantly the aphid fecundity compared with the control. Previous studies revealed that the reproductive performance of the *Drosophila melanogaster* Meigen showed variations due to Pb (Fry *et al.*, 1998) through affecting ovariole number (Wayne *et al.*, 2001). On the contrary of the fecundity, the aphid longevity was not affected by the Pb except in the case of insects frequently exposed to the Pb. Also, significant variation in the longevity of *D. melanogaster* was found by Mackay (2002) in response to the Pb.

All the Pb concentrations caused higher aphid mortalities than in the control at the both patterns of the Pb exposure. Such increase in the mortality is in line with the Parke *et al.* (1991) and Haq *et al.* (2011) findings; where the death rate of *D. melanogaster* increased with the dose of the Pb acetate. Weight decrement of the produced aphids was significant compared with the control for the 2 highest Pb concentrations when insects exposed once to Pb. When aphids frequently exposed to Pb, weights of offspring were significantly decreased from the control. The obtained weights are opposite to the expected; since El-Sheikh *et al.* (2010) found that the Pb increased the total lipid content of *Culex pipiens* L.

Many aphids, including *A. nerii*, produced winged forms in response to decreasing host plant quality (Groeters and Dingle, 1989; Dedryver *et al.*, 2008). Considering the presence of the Pb as a low-quality parameter, production of the alate offspring was higher for aphids frequently exposed to Pb-polluted leaves than for aphids exposed once and/or those exposed to the low Pb concentration. The lead polluted the plant and might make it unpalatable to insects or might cause toxicity and obliged aphid to form wings and leave the host.

The gradual increment of the Pb concentration led to a gradual prolongation of the parasitoid development period. Also, the gradual increment of the Pb concentration resulted in a decrease in the parasitoid fecundity while the parasitoid longevity was not affected. The same result of decreasing fecundity was obtained for *C. pipiens* by El-Sheikh *et al.* (2010); supposing that the significant increase in the total lipid content of the males *C. pipiens* resulted in decreasing the reproductive ability. Hirsch *et al.* (2003) found that chronic lead exposure impairs locomotor activity levels in the *Drosophila*, and this might stand behind the decreased fecundity of the parasitoid. The Pb did not cause increment in the parasitoid mortality compared with the mortality in the control, such result disagree with El-Sheikh *et al.* (2010) finding; where a concentration dependent mortality percent was obtained for *Culex*.

When the parasitoid exposed once to Pb via their larval feeding on the aphid host, only the highest Pb concentration caused sex ratio shift toward males. Moreover, gradual shift toward males was occurred due to the gradual increment of the Pb concentration at the frequent exposure to the Pb.

For many of the variables tested, it was noticed that these variables were gradually negatively affected as a result of the gradual increment in the Pb concentration, especially when insects exposed frequently to the Pb. This suggests that the Pb has an accumulative and chronic effect. Garavan *et al.* (2000) found behavioral effects of chronic Pb exposure in the invertebrates. Furthermore, results indicated that the contaminated haemolymph of the aphid (as a host for the parasitoid) with the Pb resulted in a residual effect on the parasitoid; this is supported by the finding of Timmerman and Walker (1989); that is just few insect species have the ability to eliminate metal through excretion. So that, *A. nerii* might not be able to eliminate the Pb from their body and the parasitoid was adversely affected through their larval feeding.

In conclusion, the Pb would strongly affect insects' development, fecundity, survival and the intrinsic rate of increase. Therefore, biological variables of both insects might be used as indicators for monitoring Pb pollution. The adverse effects of Pb on the studied insects were of a residual pattern; since it affected the pest insect and subsequently affected its parasitoid; therefore, Pb would negatively affect the food chain members. Moreover, this effect was more conspicuous when insects frequently exposed to Pb rather than when they were exposed once; supporting the accumulative and chronic characteristics of the Pb.

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