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Development and Reproduction of Three Predatory Mites (Acari: Laelapidae and Rhodacaridae) on Eggs of *Ephestia kuehniella* (Lepidoptera: Pyralidae)

M. F. HASSAN¹, F. M. MOMEN²*, A. K. NASR,² A. H. MABROUK¹, and M. M. RAMADAN²

¹Department of Zoology and Agriculture Nematology, Faculty of Agriculture, Cairo University, Cairo, Egypt ²Pests and Plant Protection Department, National Reseach Centre (NRC), 31 El-Bouhoth Street, 12311 Dokki, Cairo, Egypt

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The laelapid mites *Cosmolaelaps keni* and *Laelaspis astronomicus* beside the rhodacarid mite *Protogamasellopsis denticus* are biological control agents and being native predators in Egypt. In order to raise active population of above species, the biology and demographic parameters, of the three tested species were observed under laboratory conditions with eggs of *Ephestia kuhniella*. All predatory species were developed and reproduced with *E. kuhniella* eggs. The mean developmental and oviposistion periods of *L. astronomicus* females were the longest from the other tested species. The mean longevity of *C. keni* was longer than that of *P. denticus*. *Cosmolaelaps keni* consumed more *E. kuhniella* eggs during the oviposition and adult longevity periods than other species did. *Protogamasellopsis denticus* fed on *E. kuhniella* food gave the most rate of oviposition as shown by the maximum fertility, net reproductive rate and gross reproductive rate. *Laelaspis astronomicus* preying on *E. kuhniella* eggs gave the longest generation time, lowest rate of fecundity and all life table parameters compared to other tested species; therefore *E. kuhniella* eggs was appropriate for the oviposition of *P. denticus* and *C. keni* rather than *L. astronomicus*.

This study is the first to report that *E. kuhniella* eggs were an acceptable factitious food for *P. denticus*, *L. astronomicus* and *C. keni*.

Keywords: Acari, Laelapidae, Ascidae, factitious food, Pyralidae, biological control.

Laelapidae family is predators of small pests living in soil (Beaulieu, 2009). Some species of laelapid mites proved that, they may be play an important role in biological control of pest mites, insects or nematode which spends all or part of their lives in the soil (Walter and Proctor, 1999). A *Cosmolaelaps vacua* (Michael) was found to complete its development when feeding on eggs of house fly (Abou-Awad et al., 1989). *Cosmolaelaps simiplex* (Berlese) and *Cosmolaelaps qassimensis* (Fouly and Abdel-Baky) are effective biocontrol agents against the nematodes *Tylenchulus semipenetrans* (Cobb), *Meloidogyne javanica* Treub and *Meloidogyne incognita* Chitwood (Meloidogynidae) and the acarid mites, *Caloglyphus rodriguez* Samsinak and *Tyrophagus puterscentiae* (Schrank) (Al-Re-

* Corresponding author; e-mail: fatmomen@yahoo.com

hiayani and Fouly, 2005; Fouly and Abdel-Baky, 2015). Laelaspis astronomicus (Koch) is also polyphagous predator and therefore females able to feed and sustain oviposition on two species of nematodes and acarid mites (Nawar, 2011). Recently, Moreira et al. (2015) presented the importance of Cosmolaelaps jaboticabalensis Moreira, Klompen and Moraes in controlling thrips Franklinella occidentalis (Pergande) and Protorhabditis sp. Another group of predatory mites is the Rhodacaridae family, which forms frequently encountered in deeper soil layer, even into lying ground water habitat (Krantz, 1978). Lee (1970) indicated that no published data on the potential of the rhodacarids, in a strict taxonomic sense as biological control agents. Rhodacarids was observed by Sardar and Murphy (1987) to prey on small collembolans, immature oribatid mites and nematodes. Lately, two *Protogamasellopsis* sp. was cited in the literature as being predatory mites ex: Protogamasellopsis posnaniensis Wisniewski and Hirschmann was able to consume and reproduce on various prey with highly preference to T. puterscentiae, F. occidentalis and Protorhabditis sp. (Castilho et al., 2009); Protogamasellopsis zaheri Abo-Shnaf, Castilho and Moraes was able to feed and sustain oviposition on various acarid species, with the highest oviposition on Aleuroglyphus ovatus (Tropeau) and Cosmoglyphus oudemansi (Zachvatkin) (Barbosa and de Moraes, 2016).

One of the main inhibitions for the applied use of augmentative biological control in the managements of mite/insect pests is the high price of commercially produced natural enemy (De-Clercq et al., 1998). Providing solely formed unnatural food may help the mass rearing of these greedy predatory mites/insects to be more economical.

Tyrophagus puterscentiae was used as a factitious food to produce most of soil mites in large scale (Steiner et al., 1999; Freire and Moraes, 2007).

Factitious food rise the economic value of marketable production systems as predators can be reared at high bulks in a rearing component (Zhang, 2003).

Eggs of *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) are of high nutritional value (richer in protein and amino acid) and have shown to be appropriate factitious food for the mass-rearing and field releases of insect predators, ex: the mirid bugs, *Nesidiocoris tenuis* (Reuter), *Macrolophus pygmaeus* Rambur (Hemiptera: Miridae) (Molla et al., 2014) and the predatory stinkbug, *Andrallus spinidens* (F) (Heteroptera: Pentatomidae) (Mohaghegh and Amir-Maafi, 2007).

Some of phytoseiid mites (Mesostigmata: Phytoseiidae) were succeeded to complete the development and oviposited on factitious prey (*E. kuehniella* eggs) (Vantornhout et al., 2004; Momen and El-Laithy, 2007; Nguyen et al., 2014; Vangansbeke et al., 2014).

The association between laelapid and rhodacarid mites and their future factitious prey has been little reported in the literature. For example *Gaeolaelaps aculeifer* Canestrini and *Stratiolaelaps scimitus* (Womersley) succeeded to oviposit on various factitious prey including eggs of *E. kuehniella* (Navarro-Campos et al., 2016); *C. jaboticabalensis* developed and reproduced on *T. putrescentiae* as prospective factitious food (Moreira et al., 2015).

Knowledge of the biology of *Cosmolaelaps keni* Hafez, El-Badry and Nasr, *L. astronomicus* and *Protogamasellopsis denticus* (Nasr) is limited. In their research, Saber et al. (2007) misidentified the species *P. denticus* and referred it to the genus *Protogamasellus* Karg being in the family Ascidae (*Protogamasellus denticus* Nasr), presumably the above authors did not check some character's cited by Evans and Purvis (1987), accordingly some species moved from the family Ascidae to the close one Rhodacaridae. *Protogamasllopsis denticus* is among other species moved to the family Rhodacaridae based on the taxonomical criteria cited by Evans and Purvis (1987).

Here we assume that *C. keni*, *L. astronomicus* and *P. denticus* feed, oviposited and increases in great numbers on *E. kuehniella* eggs, then, selection of the best performed one which can be reared easily on that food for augmentative biological control.

So, the objective of this paper was to evaluate the relative nutritional quality of *E. kuehniella* eggs as factitious food source for those predators.

The generic concepts of the family Laelapidae follow those of Evans and Till (1979).

Materials and Methods

Host and mesostigmatic predatory mites' culture

Adult females of *C. keni* and *L. astronomicus* were obtained from soil associated with roots of tomato plants (*Lycopersicom esculentum*) L. at Tanta Governorate in February 2015. Individuals of *P. denticus* were collected in deep soil (20 cm deep) samples under banana trees (*Musa cavendishii*) L. (Family: Musaceae) at Faywam region, Egypt in 2014. Modified Tullgran funnels with 25 watt bulbs were used to extract living mites in moistened plaster-lined glass Jars. Soil samples was placed in each funnel and mites removed from collection Jars using a very fine paintbrush after 2–4 days. Different species of mites were cultured separately in plastic cells (2.5 cm in diameter and 2 cm in depth) containing a floor of plaster of Paris and charcoal (mixed in a 9: 1 ratio) were used. For biological experiments, small plastic cells (1.0 cm in diameter and 0.8 cm in depth) were used. A large heavy glass cover was used for each cell to prevent mites from escaping. Activated plaster floor kept humid by daily addition of water.

Insect eggs were added daily to each predator as food. All predatory mites was reared on *E. kuehniella* eggs for 3 months before experiments in the laboratory of (NRC), Egypt. Feeding experiments were conducted in the laboratory at 28 ± 1 °C and 70-75% relative humidity (RH). The newly deposited eggs by female's *C. keni*, *L. astronomicus* and *P. denticus* were singly removed from cultures in large cells to the small one for different biological tests.

Test diet

Fresh eggs of *E. kuehniella* were obtained daily from the Insect Department of (NRC), Egypt.

Effect of E. kuehniella *eggs on the development, consumption, reproduction and demographic parameters*

A total of 30 eggs of *C. keni*, 26 of *L. astronomicus* and 20 of *P. denticus* could be considered for the tested food, was transferred singly to the small cells. Each larva of predatory species was provided with additional of *E. kuehniella* eggs (more than that consumed daily). The number of consumed eggs was calculated daily. Fresh eggs were replaced daily.

The developmental periods of different stages of each predator were recorded every 12 h. Newly emerged females of each species were allowed to copulate with males on cells if necessary. The experimental cells were examined every 24 h to determine the duration of various physiological periods, daily consumption and oviposition rates as well as sex ratio of the progeny being calculated, too. Consumed eggs were recognized based on the occurrence of a hole or slit and a shriveled or hollow appearance. Predatory mites periodically mounted in Hoyer's medium to confirm colony identy and purity. All of the experiments were conducted under laboratory conditions of 28 ± 1 °C and 70-75% (RH).

Statistical analysis

Data presented in this paper were analyzed by one-way ANOVA and means between tested species were compared by Tukey HSD (P = 0.05 level).

Life table

The demographic parameters were calculated using Huang and Chi (2013) and their definition were those proposed by Birch (1948).

Results and Discussion

Effect of E. kuehniella eggs on development, survival and consumption rate

Larvae of all tested predatory mites are non-feeding type. Similarly, larvae of other laelapid mites ex: *C. vacua*, *C. simplex*, *S. scimitus* and *C. jaboticabalensis* did not feed (Abou-Awad et al., 1989; Al-Rehiayani and Fouly, 2005; Cabrera et al., 2005; Moreira et al., 2015).

Fresh *E. kuehniella* eggs were readily accepted by immature stages of all tested predators as factitious food. Protonymphs of *C. keni*, *L. astronomicus* and *P. denticus* developed and completed their life cycle on *E. kuehniella* eggs (Table 1). Because of the egg and larval stages of tested predatory mites are non-feeding instars, similar durations of both stages were expected in presence of insect diet. The mean development time of *L. astronomicus* from larva to adult female was significantly longer (F = 10.243, df_{2,50} p = 0.000) than that of *C. keni* and *P. denticus*. Comparing our results with other laelapid mites, indicated that: the life cycle period of *C. qassimensis*, *C. simplex* and *C. jaboti*-

cabalensis fed various nematodes ex: *M. incognita*, *M. javanica* and *T. semipenetrans* as well as the acarid mite *Caloglyphus rodriguez* Samsinak and *F. occidentalis* was twice longer than predatory laelapid mites in the present study (Al-Rehiayani and Fouly, 2005; Fouly and Abdel-Baky, 2015; Moreira et al., 2015). Results of duration of *P. denticus* life cycle were comparable to what has been mentioned by Castilho et al. (2009) for *P. posnaniensis* fed *T. putrescentiae*. The immature stages of *P. denticus* consumed a significantly lower number of *E. kuehniella* eggs (F = 23.272, df_{2,50}, *p* = 0.000) than both other nymphs of predatory mites did (Table 2). Immatures of *C. vacua* was able to consume an average of 33.0 nymphs of *Rhizoglyphus echinopus* (Fum. and Rob.) and 13.0 eggs of *Musca domestica* L. to reach the female stage (Abou-Awad et al., 1989).

Effects of E. kuehniella eggs on longevity, consumption and fecundity

In all predatory mites, male was found in each culture. The mean pre-oviposition period was likewise in *C. keni* and *L. astronomicus*. The shortest mean oviposition period (25.05 and 29.85 days) was recorded on *P. denticus* and *C. keni*, whereas *L. astronomicus* resulted in the longest mean oviposition duration (36.13 days) (F=11.258, df_{2, 50}, p=0.000) (Table 1). Insignificant difference was detected on the adult longevity of both predatory mites *L. astronomicus* and *P. denticus* (F=4.109, df_{2, 50}, p=0.022) (Table 1). A similar trend was observed for female life span of above both predators. Ahmed (1992) recorded that when the laelapid mite, *Hypoaspis zachytkinae* Sherif and Afifi fed larvae

Developmental – stages	P. denticus (18 ^z)	C. keni (20 ^z)	L. astronomicus (15 ^z)	- Calculated (F) value	Probability
Egg	$1.88\pm0.11a^{y}$	$1.85 \pm 0.13a$	1.87±0.09a	0.030 ^{ns}	0.971
Larva	1.00 ± 0.00 a	1.00 ± 0.00 a	$1.13 \pm 0.09a$	2.758 ^{ns}	0.073
Protonymph	$2.11 \pm 0.14a$	$1.90\pm0.10a$	$2.13\pm0.09a$	1.356 ^{ns}	0.267
Deutonymph	$1.94 \pm 0.09a$	$2.00\pm0.07a$	$2.73\pm0.18b$	13.124*	0.000
Life cycle	$6.94 \pm 0.15a$	$6.75\pm0.17a$	$7.87 \pm 0.21 b$	10.243*	0.000
Pre-oviposition	$1.22 \pm 0.10a$	$1.75\pm0.12b$	$2.06\pm0.22b$	7.695*	0.001
Oviposition	$25.05 \pm 1.61 \mathrm{a}$	$29.85 \pm 1.42 a$	$36.13 \pm 1.78b$	11.258*	0.000
Post-oviposition	$11.38\pm2.06\mathrm{b}$	$13.45 \pm 1.32b$	$3.27 \pm 0.24a$	11.982*	0.000
Adult longevity	37.66±2.41a	$45.05 \pm 1.24 b$	41.47±1.93ab	4.109*	0.022
Life-span	44.61±2.41a	$51.80 \pm 1.17 \text{b}$	49.33±2.10ab	3.855*	0.028

Table 1

Mean developmental periods (mean ± SE in days) of *Protogamasellopsis denticus*, *Cosmolaelaps keni* and *Laelaspis astronomicus* females fed *Ephestia kuhniella* eggs at 28±1 °C and 70±5 % R. H.

^y Means within a row followed by the same letter are not significantly different (Tukey HSD, a=0.05)

^Z Numbers between parentheses represent the number of replicates; * Significant at P = 0.05; df = 2,50; ns = non-significant

Table 2

Mean total and daily number of *Ephestia kuhniella* eggs consumed during the developmental periods and adult longevity of *Protogamasellopsis denticus*, *Cosmolaelaps keni* and *Laelaspis astronomicus* females at 28 ± 1 °C and 70 ± 5 % R. H.

Female stages	Protogamasellopsis denticus (18 ^z)		Cosmolaelaps keni (20 ^z)		Laelaps astronomicus (15 ^z)		Calculated (F)	
	Total	Daily	Total	Daily	Total	Daily	Total	Daily
Larva		Non-feeding instar						
Proto- nymph	$7.94 \pm 2.06 A^{\rm y}$	3.55 ± 0.65^a	$7.55 \pm 1.08 \mathrm{A}$	4.10±0.56a	9.73±1.03A	$4.62 \pm 0.52a$	0.561 ^{ns}	0.784ns
Deu- tonymph	$9.83\pm0.96A$	$4.97\pm0.44a$	$16.25 \pm 1.20 \text{A}$	$8.24\pm0.60b$	$25.66 \pm 2.55 \mathrm{B}$	$10.21\pm1.25b$	23.272*	11.058*
Life cycle	$17.77\pm2.75A$	$4.24\pm0.53a$	$23.80 \pm 1.84 \mathrm{A}$	$6.19\pm0.50b$	$35.40 \pm 1.77 \mathrm{B}$	$7.57\pm0.56b$	15.150*	9.430*
Pre- oviposition	$5.88 \pm 0.53 A$	$5.05\pm0.45a$	$14.80\pm2.17A$	8.56±1.03b	$29.40\pm5.20B$	13.30±1.39c	15.049*	15.950*
Oviposi- tion	361.72±22.60A	14.54±0.49a	576.90±31.75B	19.58±1.10b	463.00±19.13A	$12.95\pm0.30a$	17.859*	19.459*
Post- oviposition	263.28±49.19B	22.84±1.12b	367.35±39.70B	28.73±1.74c	25.33±1.89A	7.89±0.39a	19.104*	60.888*
Adult longevity	630.89±54.49A	$16.42 \pm 0.64b$	$959.05 \pm 22.94B$	21.37±0.32c	517.73±21.29A	$12.60 \pm 0.30a$	39.437*	90.970*
Life-span	$648.67\pm54.80\mathrm{A}$	$15.21\pm0.61b$	$982.85\pm23.03\mathrm{B}$	$20.13\pm0.26c$	553.13±20.94A	$12.06 \pm 0.31a$	38.138*	89.85*

Means within a row followed by the same upper-case letter (total consumption rate) and lower-case letter (daily consumption rate) are not significantly different (Tukey HSD, a = 0.05);

^Z Numbers between parentheses represent the number of replicates;

* Significant at P = 0.05; df = 2, 50;

ns = non-significant

of *M. domestica* and *Drosophila melanogaster* (Meigen), the duration of the adult longevity were (22.7 and 20.3 days), while Moreira et al. (2015) found a longer period of adult longevity for C. jaboticabalensis fed F. occidentalis (57.7 days). Castilho et al. (2009) indicated that the adult longevity for the rhodacarid mite *P. posnaniensis* was 68.0 days with a longer post-oviposition being 38.8 days on T. putrescentiae which approximately less than 2 and 3 times longer of the adult longevity and corresponding stage in the rhodacarid species P. denticus fed E. kuehniella eggs.

The maximum consumption rate was recorded for the adult females during the oviposition periods as *P. denticus* was consumed a total average of 361.72 *E. kuehniella* eggs, while C. keni and L. astronomicus consumed a total average of 576.90 and 463.00 eggs, respectively (Table 2). In other species of the family Laelapidae and Rhodacaridae, C. ja*boticabalensis* was able to attack 54.1 pre-pupae/pupae of *F. occidentalis* per adult predator; while P. pasnaniensis being able to consume an average of 12.8, 23.5 individuals of R. echinopus and T. puterscentiae as factitious food, with maximum laid eggs on the latter prey (Castilho et al., 2009; Moreira et al., 2015). Females of G. aculeifer consumed an average of 12.3 thrips larvae during the 3 days period, while S. scimitus attacked 8.3 individuals (Navarro-Campos et al., 2016). The present study has revealed that the mean predation rate of E. kuehniella eggs is considerably very high in C. keni, and this result support the probability that C. keni is able to feed positively and reach a very high population under certain condition. On E. kuehniella eggs, the oviposition period of C. keni was as long, its fertility was about one and half and its net reproductive rate about twice more than determined by Moreira et al. (2015) for C. jaboticabalensis fed F. occidentalis and Protorhahditis sp.

The highest mean total number of eggs deposited per female was on P. denticus, while the lowest being on L. astronomicus (F = 6346.608, df_{2 50}, p = 0.000) (Table 3). Re-

Mean total fecundity and life table parameters of Protogamasellopsis denticus, Cosmolaelaps keni and Laelaps astronomicus females, fed Ephestia kuehniella eggs at 28 ± 1 °C and 70 ± 5 % R. H.

Protogamasellopsis

Predatory mites

Cosmolaelaps keni

Table 3

	denticus (18 ^z)	(20 ^z)	astronomicus (15 ^z)	varue
Mean total fecundity (no. eggs/female)	$116.86 \pm 0.158 a^{y}$	99.94±0.320b	$78.64 \pm 0.214c$	6346.608**
Intrinsic rate of increase (r_m)	$0.33\pm0.001\text{b}$	$0.35 \pm 0.001 a$	$0.26 \pm 0.001c$	2229.808**
Net reproductive rate (R_0)	$99.63 \pm 0.922a$	$86.73 \pm 0.728b$	$65.61 \pm 0.693c$	475.741**
Mean generation time (T)	$13.86 \pm 0.24 b$	$12.82 \pm 0.29c$	$16.41\pm0.42a$	3280.637**
Finite rate of increase (λ)	$1.39\pm0.002b$	$1.42 \pm 0.002a$	$1.29 \pm 0.001c$	2164.124**
Sex ratio (females/total)	0.90	0.68	0.37	
Gross reproductive rate (GRR)	$100.98 \pm 0.921a$	87.53±0.734c	78.90 ± 0.657 b	204.338**

^y Means within a row followed by the same letter are not significantly different (Tukey HSD, a= 0.05)

^Z Numbers between parentheses represent the number of replicates;

**Highly significant at p = 0.000

Life table parameters

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Laelaspis

- Calculated (F)

value

search has been done by Castilho et al. (2009) indicated that the total fecundity of *P. pos-naniensis* fed *T. puterscentiae* was (106.6 eggs/female), which being close to the rhodacarid mite fed *E. kuehniella* eggs in the present study. Many species of mites of the family Phytoseiidae, it has been proved that the rate of oviposition is completely correlated with the predation rate (Momen, 1996; Momen and El-Borolossy, 1999), because plenty of the material eaten by female mites is adapted into predator eggs.

Effect of E. kuehniella eggs on demographic parameters

With *E. kuehniella* eggs, the net reproductive rate (Ro) was significantly higher (99.63 and 86.73) on *P. denticus* and *C. keni*, than on *L. astronomicus* (65.61) (Table 3). The intrinsic rate of natural increase (r_m) and consequently, the finite rates of increase (λ) were significantly higher on *C. keni* (0.35 and 1.42) than on *P. denticus* (0.33 and 1.39) or *L. astronomicus* (0.26 and 1.29). The mean generation time (T = 16.41 days) was significantly longer on *L. astronomicus*, than on *P. denticus* (13.86 days) or *C. keni* (12.82 days). The gross reproductive rate (GRR) was significantly higher in *P. denticus* than on *C. keni* and *L. astronomicus*. The high value of (λ) in *C. keni* could be results from both a short value of (T), which is important and relatively a higher than those reported by Moreira et al. (2015) for *C. jaboticabalensis* fed *F. occidentalis* and *T. puterscentiae* as well as *C. simpex* fed *C. rodriguezi* and *M. Javanica* (Al-Rehiayani and Fouly, 2005). This difference may be due to the food types and different mites studied.

Our study is the first record of the life history for *P. denticus*, *C. keni* and *L. astronomicus* fed *E. kuehniella* eggs as a factitious food, and also explained that the tested food being clearly accepted as unnatural food to those predatory mites.

Because, no much data on the real type of food substances that tested predatory mites use in the field, much more work is essential to define what items of food be used.

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