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Energy reserves in *Brevicoryne brassicae* (Lin.) (Homoptera: Aphididae) instars and their effect on predation and longevity of three coccinellid species under laboratory conditions

Salama Ibrahim Askar¹ and Monir Mohamed El Husseini^{2*} 

Abstract

Background: Contents of carbohydrates, protein, and lipids in tissues of insect prey constitute the required energy reserve utilized by their predators. Their amounts define the preference of the predator to certain prey on which its biological performance is the optimum. Feeding preferences and longevity of the 3 coccinellid beetles; *Coccinella septempunctata* L., *C. undecimpunctata* L., and *Scymnus interruptus* L. were studied in relation to energy reserve in tissues of the cabbage aphid, *Brevicoryne brassicae* (Lin.).

Main body: The level of energy reserve increased with increasing of aphid nymphal instars showing the highest in 4th instar (0.10844 cal/mg). Lipids and carbohydrates represented the highest energy reserve average rate in *B. brassicae* adults (0.29139 and 0.07404 cal/mg), respectively. *C. septempunctata* larval stage recorded the highest efficiency in daily predation (89.42 prey), when fed on 1st instar nymphs of the aphid. It required 21 days when fed on the 3rd nymphal instar of *B. brassicae*. *S. interruptus* larval stage reflected the lowest consumption and longevity in predation where it consumed less than 69.25 prey/day and lasted a period less than 17.5 days. Generation time for *C. undecimpunctata* lasted the longest (20.89 days) when fed on *B. brassicae* 1st nymphal instar. It was decreased in *S. interruptus* recording (12.22 days) when fed on *B. brassicae* 3rd nymphal instar.

Conclusion: Only *C. septempunctata* was efficient in predation and its biological performance was the best compared to the other tested coccinellid species. Use of *C. septempunctata* can be recommended when applying biological control programs in the field against the cabbage aphid.

Keywords: Energy reserves, *Brevicoryne brassicae*, Coccinellidae, Predation

* Correspondence: monir.elhusseini@agr.cu.edu.eg

²Center of Biological Control & IPM, Faculty of Agriculture, Cairo University, Giza, Egypt

Full list of author information is available at the end of the article



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Background

Predators have a great potential to keep the population of aphid under control (Askar et al. 2013). The coccinellids represent an important group of predators that manage pests' populations throughout the world (Obrycki and Kring 1998). Naturally, coccinellids feed on aphids, thrips, spider mites, and various soft-bodied insects and their eggs. In aphids, many factors can affect the prey preferences of the coccinellids, including the mobility and defense reaction of the prey (Provost et al. 2006), morphological characters and previous feeding experience (Khan and Rafique 2004), predator and prey size (Thompson 1975), and abundance of prey (Soares et al. 2004). Many species of aphids are characterized by production of some biological properties such as susceptibility to natural enemies (Brandele and Weisser 2001). Information pertaining to the biological parameters and feeding preference of predatory beetle on different aphid instars is essential for assessing the potential rate of increase in the population and its predation as a biological control agent.

The aim of this study was to determine the effect of energy reserves of *Brevicoryne brassicae* (L.) instars on feeding preferences and longevity of 3 coccinellid beetles as well as measuring the energy reserves quantitatively including water-soluble carbohydrates, lipids and proteins in different instars of *B. brassicae* under laboratory conditions.

Main text

Material and methods

Cabbage plants were cultivated in plastic pots (25 cm diameter, 20 cm high), kept in net cages of 50 × 60 × 60 cm under a climate-controlled room of 25 ± 2 °C, 65 ± 5% RH and a 14L:10D photoperiod. One-month-old plants, with 5–7 leaves, were used for rearing the cabbage aphid species, *B. brassicae*.

Source of the aphid and the coccinellid species

Infested plants with the cabbage aphid, *B. brassicae* infestation were collected from cultivated cabbage fields in El-Behera Governorate, Egypt. The collected aphids were colonized in the laboratory on cultivated cabbage plants and kept at 25 ± 2 °C, 65 ± 5% RH and 14L:10D photoperiod. Under the same laboratory conditions, the selected 3 coccinellid predators, *Coccinella septempunctata* L., *C. undecimpunctata* L., and *Scymnus interruptus* L. were collected from the field, reared, and colonized on cabbage aphid. They were reared for one generation before their immature and mature feeding individuals being used in the experimental study.

Quantitative measuring of the energy reserves

To determine the different energy reserves (lipids, proteins, and carbohydrates), *B. brassicae* adults and

nymphal instars were selected randomly, then separately homogenized for 30 s in 180 µl of aqueous lysis buffer solution (100 mM KH₂PO₄, 1 mM dithiothreitol, and 1 mM ethylenediamine tetra acetic acid, pH 7.4), using a plastic micro-pestle (Lowry et al. 1951). Three replicates of each determination in the following assays were analyzed. According to Lowry et al. (1951), protein concentrations in homogenates of the whole body of the aphid instars were measured. Bovine serum albumin at appropriate concentrations was used as the standard. Total carbohydrates were dissolved by addition of 20 µl of sodium sulfate solution (20%) to 180 homogenates (Van Handel and Day 1988). Total lipid and water-soluble carbohydrates were solubilized by mixing the solution with 1500 µL of a chloroform-methanol solution (1:2 v/v) (Van Handel and Day 1988). Each sample was then centrifuged at 16,000 rpm for 15 min at 4 °C. The supernatant was transferred to a new micro-tube for water-soluble carbohydrate determination. The pellet was used for the determination of the glycogen content. Vanillin reagent and cholesterol (as the standard) were used for measuring total lipid content (Van Handel 1985). One hundred microliters of the supernatant from centrifuged chloroform-methanol solution was transferred into new micro-tubes and heated until complete solvent evaporation. Ten microliters of 98% euphoric acid was added to each micro-tube, which incubated at 90 °C for 2 min. Vanillin reagent (190 µl) was added to each ice pre-cooled micro-tube. Absorbance was determined at 540 nm after 15 min incubation at room temperature. Different energy reserves for lipids, proteins, and carbohydrates were converted into energetic equivalents according to Ahsaei et al. (2013). Feeding performance and longevity of the coccinellid species were defined.

Predation efficiency and longevity of the coccinellid beetles

To estimate some biological parameters, prey consumption, longevity of the 3 coccinellid species, and the effect of aphid prey instars on these measures were performed. The experiments were conducted, using separately the second generation of each predatory species. Cabbage leaves were cut into pieces and placed in glass Petri dishes (9-cm diameter) containing 2% agar at the bottom. Singly, each aphid nymphal instar was counted and transferred into a glass Petri dish to feed on cabbage leaves. After hatching of the predatory species, larvae were collected with a fine camel hairbrush and placed into a new glass jar. Each larval instar was provided daily by a known number of aphids (at least 150 individual prey) at different adult and nymphal instars for feeding the predator. During development of larval instars and adults, remaining number of aphids were daily counted and offered other known number to give each predator instar enough prey number till developing to the next

instar. Feeding potential was recorded by counting the number of aphids consumed by 1st, 2nd, 3rd, and 4th instars for the tested coccinellids up till the adult started to lay eggs. Five replicates for each treatment were conducted, each with 5 larvae. Aphids' prey was also provided by enough numbers of the same instar for each predatory instar. Control units were free from any predator and were also inspected daily to record the increase of the progeny or of the mortality of aphid prey in order to correct the number of actual consumed aphids in each experiment. Data were collected twice daily (each 12 h) and corrected with that of the control (Abbott 1925) to evaluate feeding capacity and longevity of each coccinellid instar under the influence of different instars of aphid prey. Experiments using the same design were conducted with *C. undecimpunctata* and *S. inter-ruptus* adults and larval instars.

Statistical analysis

Data were analyzed using (SAS Institute 1988) to test the significance and estimate LSD among treatments. The correlation relationships and correlation coefficient values between predator and prey age, was estimated as the regression line relationship was calculated by the regression line equation.

Results and discussion

Energy reserves in *B. brassicae* nymphal instars

The energy reserves encountered the carbohydrates, proteins and lipids in *B. brassicae* nymphal instars. The average energy reserves in instars differed significantly ($P > 0.05$) (Table 1). The level of portions increased by increasing age of the nymphal instar, recording the highest in 4th instar (0.10844 cal/mg). The results also showed that the energy presented in the form of lipids and carbohydrates increased by the advancement of the nymph age, till they recorded the highest average rate in *B. brassicae* adults with (0.29139 and 0.07404 cal/mg), respectively. The general energy level increased as the nymphs advanced in age to record the highest in *B.*

brassicae adults with 0.473839 cal/mg. In contrast, the average protein energy reserves in *B. brassicae* aphid over their lifetime was lower than the general average level of lipids, but at the same time it was higher than the one of carbohydrates, where the overall average scored (0.375227 cal/mg) for all ages and during the life cycle of aphids (Table 1). Results also showed that a convergence in the energy level between the 4th nymphal instars and adults, other a convergence between the 2nd and 3rd nymphal instars. The results reflected that lipids were the most abundant energy reserve (63.67%), followed by proteins (24.15%) and then soluble carbohydrates (12.18%). The results also revealed that protein and lipids were the most important storage fuels of insects and were utilized more frequently than other reserves to obtain energy for muscular activity, so there were significantly higher contents of lipids, but lower contents of proteins and soluble carbohydrates. Obtained data are nearly in accordance to those of Ahsaei et al. (2013) who found in the pea aphid, *Acyrtosiphon pisum*, a higher percentage content of water-soluble carbohydrates and lipids in the red clones. Thus, this aphid species needs more lipids and carbohydrates to fuel their movements and flight. Gupta and Kumar (2017) reported that protein concentration values were directly proportional to the efficiency of artificial diets for *C. septempunctata* larvae.

Predation efficiency and longevity of *C. septempunctata*

Data presented in Table 2 showed longevity and feeding consumption of *C. septempunctata* different stages, when fed on different instars of *B. brassicae*. Number of consumed prey increased gradually as the age of predator and the prey increased. The predator lifespan had non-significant effect on different ages of prey (Table 2). The 1st instar larvae of *C. septempunctata* consumed a highest number of 1st instar of the aphid (42.33 daily). Feeding efficiency decreased gradually as well as the prey increased in age. On other hand, 2nd and 3rd instars showed non-significant variations among all prey instars.

Table 1 Average energy equivalents (cal/mg) of the proteins, lipids and carbohydrates in *B. brassicae* instars

Individual <i>B. brassicae</i> instar	Proteins		Lipids		Carbohydrates		Total energy
	(cal/mg)	%	(cal/mg)	%	(cal/mg)	%	
1st	0.05685 a	22.91	0.17006 a	68.53	0.02126 a	8.57	0.248161 a
2nd	0.08622 b	25.33	0.21807 b	64.07	0.03607 b	10.60	0.340355 b
3rd	0.10318 bc	26.21	0.24577 b	62.44	0.04466 b	11.35	0.393617 c
4th	0.10844 c	25.21	0.26918 bc	62.58	0.05255 c	12.22	0.430161 d
Adult	0.0984b c	21.22	0.29139 c	62.82	0.07404 d	15.96	0.473839 d
Mean	0.09062	24.15	0.23889	63.67	0.04572	12.18	0.375227
LSD 5%	0.04221	2.09	0.04913	2.79	0.06022	1.92	0.05611

Table 2 Effect of *B. brassicae* stages on *C. septempunctata* biological parameters

<i>B. brassicae</i> instar	<i>C. septempunctata</i> instars													
	1st		2nd		3rd		4th		Pupa		Adult		Pre-oviposition	
	Long.*	Prey**	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey
1st	3.17 bc	42.33 a	5.17 b	82.00 a	6.83 a	96.67 a	8.67 a	136.67 a	6.17 b	0.00	28.67 ab	167.00 a	3.17 a	167.00 a
2nd	4.17 a	34.33 b	5.50 ab	69.67 b	6.83 a	77.00 b	7.83 bc	132.33 a	6.17 b	0.00	29.33 ab	154.33 ab	3.17 a	154.33 ab
3rd	2.83 c	25.33 c	4.67 c	61.67 c	6.17 b	59.67 c	7.33 c	123.00 ab	5.83 b	0.00	27.67 b	143.67 b	2.83 a	143.67 b
4th	3.33 b	24.33 cd	5.67 a	57.33 c	6.17 b	56.67 c	8.17 ab	114.33 bc	7.00 a	0.00	28.67 ab	126.67 c	3.17 a	126.67 c
Adult	2.83 c	19.67 d	5.67 a	59.00 c	6.67 ab	54.00 c	8.67 a	107.33	6.83 a	0.00	32.33 a	118.33 c	3.33 a	118.33 c
LSD 5%	0.42	0.09	0.49	4.79	0.60	12.92	0.56	14.17c	0.62	0.00	4.38	14.09	0.52	14.09

*Longg. = longevity (/days), **Prey = number of consumed prey

Fourth larval instar consumed a high number of 1st and 2nd aphid instars (136.67 and 132.33) individuals daily, respectively. Meanwhile, feeding consumption decreased gradually recording (107.33), when fed on aphid adults. Before starting egg laying, *C. septempunctata* consumed all the aphid instars with a highest capacity on 1st nymphal instar of 167.00 that decreased gradually in 2nd, 3rd, 4th instars and adults, respectively. Concerning lifespan and longevity of *C. septempunctata*, the 1st larval instar lasted an average period of 4.17 days, when fed on *B. brassicae* instars. This period decreased in the 2nd instar under the same conditions recording (4.67 to 5.67 days). Aphid prey showed non-significant effect on the 3rd instar lifespan, but a strong effect on 4th instar duration. Meanwhile, *C. septempunctata* larvae fed on aphid from 1st to 3rd instar and showed a significant effect on pupal longevity, averaged 6 days. This period increased when the predator was fed on 4th instar and adults. Aphid instars were offered daily to adult predator till their death recording a high significant effect; adult predators lived longer time between 27.67 and 32.33 days. In the case of adult, predator pre-ovipositional period was never affected by prey instar showing a time ranged between 2.83 and 3.33 days. Singh and Singh (2013) revealed that the 1st, 2nd, 3rd, and 4th instar larvae of *C. septempunctata* consumed 21.62, 48.03, 73.37, and 102.64 aphids/day, respectively, where the feeding potential of *C. septempunctata* increased by increasing age of the larvae. Rauf et al. (2013) reported that *C. septempunctata* showed significant differences with respect to all temperatures, predatory potential of 1st, 2nd, 3rd, and 4th larval instars from 21.0 to 37.0, 40.5 to 79.0, 73.8 to 124.8, and 342.1 to 481.5 aphids, when adult male and female longevity of *C. septempunctata* ranged from 30.0 to 44.7 and 43.7 to 60.3 days, respectively. Manpoong et al. (2016) recorded that a single *C. septempunctata* adult consumed an average of 81.55 ± 15.34 aphids per day. The total larval period was observed to be 26 ± 3 days. The pupal period was found to be 7.5 ± 1.5 days. The adult female lived for 31.5 ± 1.5 days and laid 357.45 ± 22.41 eggs in her lifetime. Mahyoub et al.

(2013) recorded that *C. septempunctata* total developmental period occupied from 16 to 21 days under the constant conditions of 23 ± 2 °C and 60 ± 5 RH%.

Predation efficiency and longevity of *C. undecimpunctata*

First larval instar of *C. undecimpunctata* did not show a significant difference in the rate of predation on the first 3 instars of aphid nymphs, also between the 4th nymphal instar and adult. *C. undecimpunctata* 1st larval instar lasted a period ranged between 3 and 4 days. However, the 2nd larval instar showed a high feeding capacity on 1st and 2nd aphid nymphs, then decreased significantly on the 3rd and 4th prey nymph and aphid adult, respectively. *C. undecimpunctata* 2nd larval instar lasted an average period of feeding on the different aphid instars from 4.33 to 5.67 days as shown in Table 3. Meanwhile, *C. undecimpunctata* 3rd larval instar showed significant differences in predation rate on all of aphid's instars (Table 3). It also showed a longer lifespan with significant differences, when feeding on the different aphid instars and recorded an average lifetime ranged between 6.17 and 7.52 days. The 4th larval instar of *C. undecimpunctata* consumed 131 prey of 1st aphid nymphal instar per day. This rate decreased gradually as consumed only 83 prey of aphid adults. *C. undecimpunctata* 4th larval instar recorded a period ranged between 5 and 5.33 days to complete development. The predator pupal stage completed its development in a period between 4.33 and 5.5 days. *C. undecimpunctata* adult showed its predatory efficacy at a minimum rate of 107.33 prey/day at a minimum period of 34.33 day. Meanwhile, adult predator showed a pre-ovipositional period ranged from 4.17 to 5 days. It was always noticed that *C. undecimpunctata* fed on aphid instars, had consumed a number of prey increased by the increase of predator's age, and decreased by the increase of prey's age. Generally, the adult predator recorded the highest predatory efficacy on the 1st aphid instar, while the 1st larval instar of the predator recorded the lowest predation rate on aphid adult. Accordingly, the daily predation rate of *C. undecimpunctata* ranged between 14.33 and 140.33 prey/day

Table 3 Effect of *B. brassicae* instar on *C. undecimpunctata* biological parameters

<i>B. brassicae</i> instar	<i>C. septempunctata</i> instars													
	1st		2nd		3rd		4th		Pupa		Adult		Pre-oviposition	
	Long.*	Prey**	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey
1st	4.00 a	25.67 a	5.00 ab	75.00 a	7.52 a	95.00 a	5.17 a	131.67 a	5.17 a	0.00	36.33 a	136.00 ab	4.33 ab	136.00 ab
2nd	4.17 a	22.00 a	5.17 a	64.67 a	6.83 ab	86.00 b	5.33 a	122.67 ab	5.33 a	0.00	35.67 a	140.33 a	4.83 ab	140.33 a
3rd	3.67 ab	23.33 a	5.17 a	46.67 b	7.00 a	83.00 bc	5.00 a	120.33 b	4.33 b	0.00	34.33 a	124.67 b	4.17 b	124.67 b
4th	3.00 b	15.00 b	5.67 a	40.33 b	6.33 bc	81.33 bc	5.00	100.33 c	5.33 a	0.00	34.33 a	112.67 c	4.83 ab	112.67 c
Adult	3.67 ab	14.33 b	4.33 b	46.00 b	6.17 c	75.00 c	17 a	83.00 d	5.50 a	0.00	37.33 a	107.33 c	5.00 a	107.33 c
LSD 5%	0.79	4.53	0.74	15.34	0.65	8.12	5.33 a	9.70	0.53	0.00	3.45	11.95	0.68	11.95

*Long. = longevity (/days), **Prey = number of consumed prey

during its life time. Preferential feeding response could be affected by aphid's species according to Mari et al. (2016) who reported that preferential feeding response and instar durations of *C. undecimpunctata* when reared on 3 aphid species, i.e., *Rhopalosiphum maidis* (Maize aphid), *Therioaphis trifolii* (Alfalfa aphid), and *Lipaphis erysimi* (Mustard aphid). The duration values in days for 1st, 2nd, 3rd, and 4th instars ranged between 6.50, 5.60, 7.90, and 8.01, respectively, for *R. maidis*. For *T. trifolii*, it ranged between 5.25, 6.0, 6.10, and 7.80, respectively. Meanwhile, it ranged between 5.23, 5.66, 5.91, and 8.50 days, respectively, for *L. erysimi*. Female adult beetle lifespan under this study was 35, 50, and 40 days, while it was 30, 42, and 32 days for male adult on maize, alfalfa, and mustard aphids, respectively. They also found that the 1st instar of the predatory beetle devoured 5.50, 10.95, and 6.5 maize, alfalfa, and mustard aphids, respectively; 2nd instar devoured 20.01, 36.11, and 25.5; the preferential feeding response of 3rd instar larvae was 30.12, 46.5, and 36.5; and 4th instar larvae devoured 41.25, 57.12, and 46.11 maize, alfalfa, and mustard aphids, respectively.

Rajput (1994) reported that under laboratory conditions (about 25 °C), *C. undecimpunctata* required 17 days to reach adult stage and at 18 °C, it needed 23.6 days. Hodek (1970) reported that pupal period ranged from 4 to 6 days, with an average of 4.7 days for *C. septempunctata*. Rodriguez-Soana and Miller (1995)

reported that females and males of *C. septempunctata* lived an average of 47.6 and 46.9 days, respectively, at 20 °C. Singh and Marwaha (2002) reported that *C. undecimpunctata* grubs can consume 79 *Aphis craccivora* in a day. Manpoong et al. (2016) recorded that mean consumption of aphids per *C. undecimpunctata* adult was 80.8 individuals, whereas, 21.76, 55.67, 107.86, and 231.03 aphids were consumed by a single larva during 1st, 2nd, 3rd and 4th instars, respectively.

Predation efficiency and longevity of *S. interruptus*

The results shown in Table 4 revealed that the 1st larval instar of *S. interruptus* consumed a maximum of 20.67 prey/day, and non-significant difference recorded in its predation on all aphid instars. Also, a significant difference in the duration of its lifespan appeared, when fed on different ages of aphids, as it ranged between 2.83 and 3.17 days. On the other side, the predation rate of the 2nd larval instar of the predator increased to record the highest predation rate on the 1st prey instar at a rate of 71 prey/day through a period of 3.17 days before molting to the 3rd instar. The 3rd larval instar of the predator did not significantly exceed the daily predation rate of the 2nd larval instar registering its highest level of 84 prey individuals from the first perennial age per day and also lived a period ranged between 5 and 6.17 days to complete its life. The 4th larval instar recorded the highest predatory level, and non-significant

Table 4 Effect of *B. brassicae* stages on *S. interruptus* biological parameters

<i>B. brassicae</i> instar	<i>S. interruptus</i>													
	1st		2nd		3rd		4th		Pupa		Adult		Pre-oviposition	
	Long.*	Prey**	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey	Long.	Prey
1st	2.83 ab	20.67 a	3.17 a	71.00 a	6.17 a	84.00 a	5.00 a	101.33 a	5.33 ab	0.00	21.67 a	105.00 a	3.67 c	105.00 a
2nd	2.50 b	19.33 a	3.17 a	58.00 b	5.67 abc	80.33 ab	5.00 a	98.00 a	5.00 c	0.00	17.67 bc	94.00 b	4.50 ab	94.00 b
3rd	2.50 b	14.00 c	3.00 a	48.67 c	5.00 c	75.67 abc	4.83 a	93.33 a	4.00 d	0.00	17.33 c	91.00 b	3.33 c	91.00 b
4th	2.83 ab	18.00 ab	3.33 a	43.33 c	5.33 bc	67.67 c	5.17 a	76.00 b	5.17 bc	0.00	18.00 bc	79.33 c	4.17 b	79.33 c
Adult	3.17 a	15.33 bc	3.17 a	44.67 c	5.83 ab	70.67 bc	5.33 a	76.33 b	5.50 a	0.00	19.00 b	73.00 c	4.67 a	73.00 c
LSD 5%	0.45	3.78	0.44	7.20	0.78	10.61	0.80	10.68	0.32	0.00	1.38	10.76	0.49	10.76

*Long. = longevity (/days), **Prey = number of consumed prey

Table 5 Consumption and duration (/days) of three predatory larval stages in relation to prey energy reserve

<i>B. brassicae</i> instars	Total energy, cal/mg	Total larval instars					
		<i>C. septempunctata</i>		<i>C. undecimpunctata</i>		<i>S. interruptus</i>	
		Duration	Prey*	Duration	Prey*	Duration	Prey*
1st	0.248161 a	23.83 a	89.42 a	21.17 ab	81.83 a	17.17 a	69.25 a
2nd	0.340355 b	24.33 a	78.33 b	21.50 a	73.83 b	16.33 ab	63.92 a
3rd	0.393617 c	21.00 b	67.42 c	20.83 ab	68.18 b	15.33 b	57.92 b
4th	0.430161 d	23.33 a	63.17c	20.17 bc	59.25 c	16.67 a	51.25 c
Adult	0.463839 d	23.83 a	60.00 d	19.50 c	54.58 c	17.50 a	51.75 c
Instar LSD 5%		1.32	5.89	1.32	6.32	1.22	5.93
Regression Equation		No. of consumed prey = - 42.07 + 5.76*		No. of Consumed prey = - 40.19 + 4.09*		No. of consumed prey = - 30.30 + 5.98*	
<i>r</i>		0.97		0.29		0.98	
<i>R</i> ²		0.93		0.081		0.96	

*Prey = number of consumed prey

difference was shown when fed on the 1st, 2nd, and 3rd instar nymphs of the aphid. The results showed that predation rate of adults (73 prey) was at a level close to the daily predation rate of its 4th larval instar (76.33 prey). Also, the adult longevity ranged between 17.33 and 21.67 days in relation to consumed prey instar; the pre-ovipositional period accordingly ranged between 3.33 and 4.67 days (Table 4).

Effect of prey energy reserve on larval consumption and duration

Results for larval predation efficiency and duration of the 3 studied coccinellid larval species are presented on Table 5. Results showed non-significant deference in the total energy reserves between 4th nymphal instar and adults of *B. brassicae* as prey. Accordingly, the 3 predators, *C. undecimpunctata*, *C. septempunctata*, and *S. interruptus*, did not show a significant difference in feeding consumption on the 4th nymphal instar and *B. brassicae* adults. Moreover, the average longevity period of the 3 predatory larval stages decreased by increasing the total energy reserves. *C. septempunctata* larval stage recorded the highest efficiency in daily predation (89.42 prey), when fed on 1st aphid nymphal instar. On the other hand, its larval stage required 21 days, when fed on 3rd nymphal instar of *B. brassicae*. *S. interruptus* larval stage showed the lowest predation activity (69.25 prey/day) and longevity (17.5 days). The lowest correlation value for the 2nd predator recorded a significant difference between the predators *C. septempunctata* and *C. undecimpunctata*, and the predation rate was indicated by a decrease in prey age represented by an inverse relationship. Singh and Singh (2013) reported that larvae of *C. septempunctata* preyed on high numbers of mustard aphids (average 61.42/day) during their larval span. Singh and Singh (2014) also reported average larval

duration of *C. septempunctata* was 11.15 days on mustard aphid. Average pupal period was 5.8 ± 0.91 days. The adult male and female survived for 14–18 days and 18–24 days, respectively. Omkar and Srivastava (2003) observed *C. septempunctata* with the highest and rapid larval development (14.2 days) and greatest daily consumption of mustard aphid (45.3/day). Obtained results are also in accordance with those of Mari et al. (2016), the statistical analysis showed a highly significant difference among predatory efficiency of *C. septempunctata* instars. Imam (2015) recorded that *C. septempunctata* total larval and pupal durations were 23.4 ± 0.35 and 5.3 ± 0.56 days, respectively. Singh and Singh (2014) also reported average grub duration was 11.15 days on aphid. Average pupal period was 5.8 ± 0.91 days. The adult male and female survived for 14–18 and 18–24 days, respectively. Omkar and Srivastava (2003) observed the *C. septempunctata* with a highest rapid larval development (14.2 days) and greatest daily consumption of mustard aphid at the rate of 45.3 aphids/day. Obtained results and according to Mari et al. (2016), statistical analysis showed a highly significant difference between predatory efficiency of *C. septempunctata* larval stage (*F* = 123.15 *df* = 3, *P* < 0.05). Manpoong et al. (2016) recorded that *C. undecimpunctata* total larval and pupal durations were 23.4 ± 0.35 and 5.3 ± 0.56 days, respectively.

Generation time

Results of generation time of the 3 coccinellid species are presented in Table 6. The results showed that *C. undecimpunctata* required the longest generation time (20.89 days), when fed on *B. brassicae* 1st nymphal instar. Generation time decreased in case of *S. interruptus* recording (12.22 days), when fed on *B. brassicae* 3rd instar. Sarwar and Saqib (2010) reported that both larvae and adults of *C. septempunctata* when fed on aphid and

Table 6 Feeding consumption and generation period of three coccinellid species reared on different stages of *B. brassicae*

Coccinellid spp.	<i>B. brassicae</i> instars									
	1st		2nd		3rd		4th		Adult	
	Gener./day*	Prey/day**	Gener./day	Prey/day	Gener./day	Prey/day	Gener./day	Prey/day	Gener./day	Prey/day
<i>C. septempunctata</i>	19.56 b	85.47 a	19.94 b	77.56 a	18.17 b	70.36 b	19.67 a	63.28 a	21.00 a	59.44 a
<i>C. undecimpunctata</i>	20.89 a	72.61 b	20.83 a	71.39 a	19.83 a	64.11 a	19.94 a	58.00 a	20.78 a	53.97 a
<i>S. interruptus</i>	14.72 c	58.08 c	13.00 c	52.64 b	12.22 c	49.64 c	13.28 b	43.53 b	14.00 b	41.58 b
LSD 5%	1.16	5.65	0.82	8.97	0.96	6.07	0.91	6.12	2.06	5.77

*Gener./day = generation/day; **Prey/day = number of consumed prey/day

artificial diet showed normal development from egg to adulthood in 20.6 days on aphid prey.33

Conclusion

Results revealed the effect of prey energy reserve contents (cal/mg), available as carbohydrates, protein, and lipids in tissues of the cabbage aphid *B. brassicae*, utilized as prey for 3 coccinellid predators. Different prey nymphal instars and adults affected certain biological parameters of the predators due to their contents of energy reserve. The results reflected that *C. septempunctata* was the most efficient in predation.

Abbreviations

cal: Calories; Gener.: Generation; Long.: Longevity; r: Regression equation

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Cover letter

We did intend to submit this research paper to *The Egyptian Journal of Biological Pest Control* and it is not currently under consideration elsewhere.

Authors' contributions

SIA analyzed and interpreted the data regarding the energy reserve consumed by each predator and shared in writing the manuscript. MMEH performed the laboratory experimentation of the three predatory species and was a major contributor in writing the manuscript. All authors read and approved the final manuscript

Authors' information

Salama Ibrahim Askar is Ass. Prof. at Plant Protection Department, Faculty of Agriculture, Damanhour University, Egypt.

M.M. El Husseini (CA) is Director of the Center of Biological Control & IPM at the Faculty of Agriculture, Cairo University, Giza, Egypt.

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Author details

¹Plant Protection Department, Faculty of Agriculture, Damanhour University, Damanhour, Egypt. ²Center of Biological Control & IPM, Faculty of Agriculture, Cairo University, Giza, Egypt.

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