

Suitability of different artificial diets for development and survival of stages of the predaceous ladybird beetle *Eriopis connexa*

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Abstract The ladybug *Eriopis connexa* (Germar) (Coleoptera: Coccinellidae) is an important natural enemy of various pests. The potential of rearing it on 17 different diets was evaluated. The percentage of *E. connexa* adults was higher when its larvae received only eggs of *Anagasta kuehniella* (Zeller) (Lepidoptera: Pyralidae) after freezing for 1 day (92.5%) or com-

bined in artificial diets with honey and water (82.5% to 100.0%). The viability from larvae to adult was 72.5% with eggs of *A. kuehniella* (after 1 day's freezing) plus an artificial diet based on pet food. No adults of *E. connexa* were obtained with artificial diets as a stand-alone food source. The duration of the larval period to adult of this predator was longer, but with low viability, with only *A. kuehniella* eggs (after 6 months' freezing) or with eggs + artificial diets. Eggs of *A. kuehniella* (after 1 day's freezing) supplied separately or along with artificial diets were more appropriate to rear *E. connexa* and both diets can be used for mass rearing of this natural enemy.

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Introduction

Food quality and choice has a direct impact on the growth, development and reproduction of insects, including predators (Omkar et al. 1997, 2006; Omkar and Pervez 2001; Zanuncio et al. 2002). Low-cost high-quality foods can reduce the costs of producing predators and stimulate their use in biological control programs (Saavedra et al. 1996; Vandekerckhove et al. 2006).

Coccinellidae (Coleoptera) species are important biological control agents, because they may feed on

aphids, aleyrodids, cochineals, mites, eggs and on larvae of Lepidoptera and Coleoptera (Omkar and Pervez 2001; Ragkou et al. 2004; Silva and Bonani 2008; Simmonds et al. 2000). It is necessary to understand the nutritional and ecological patterns in order to increase the efficiency of entomophagous insects (Mohaghegh and Amir-Maafi 2007; Pervez and Omkar 2004; Simmonds et al. 2000; Zanuncio et al. 1996). Knowledge of the biology, behavior and rearing procedures are important to improve the use of predatory insects in biological control programs; the production of adequate natural diets represents a problem to mass rearing Coccinellidae that feed on aphids (Kariluoto 1980; Kariluoto et al. 1976).

An appropriate artificial diet is nutritionally comparable to a natural diet but with a lower cost. On the other hand, insects reared on an artificial diet should exhibit high survival rate, shorter development and high weight gain and fecundity (Dong et al. 2001; Saavedra et al. 1997; Scriber and Slansky 1981; Zanuncio et al. 1996). Artificial diets using mammal liver and vitamin supplements have been developed for the predator *Coleomegilla maculata* (DeGeer) (Coleoptera: Coccinellidae; Attallah and Newson 1966). The use of fresh liver of pig has been described to rear the coccinellids *Adalia bipunctata* (L.), *Coccinella septempunctata* (L.), *Coccinella transversoguttata richardsoni* (Brown), *Hippodamia tredecimpunctata tibialis* (Say) and *Propylea quatuordecimpunctata* (L.) (Kariluoto 1980; Kariluoto et al. 1976). Besides, coccinellids are reared on semi-defined diets lacking meat, but usually complemented with prey (Attallah and Newson 1966; Kariluoto et al. 1976; Matsuka et al. 1982). Factitious prey such as eggs of *Anagasta kuehniella* (Zeller) (Lepidoptera: Pyralidae) are suitable prey for rearing Coccinellidae in place of natural prey (De Clercq et al. 2005; Kato et al. 1999a, b; Specty et al. 2003).

Eriopis connexa (Germar) (Coleoptera: Coccinellidae) feeds on aphids in South America (Gyenge et al. 1998) and its mass rearing can be useful for biological control programs. Mass rearing *E. connexa* was tested with the prey *Diuraphis noxia* (Mordvilko), *Myzus persicae* (Sulzer) and *Acyrtosiphon pisum* (Harris) (Hemiptera: Aphididae) at 30°C and 34°C; results were best with the first prey at 34°C (Miller and Paustian 1992). The objective of this work was to evaluate the development of *E. connexa* on 17 different diets in an attempt to contribute to mass rearing this predator under laboratory conditions.

Material and methods

The experiment was carried out at 25±1°C, 12 h photophase and 70±10% relative humidity in the Laboratory of Insect Rearing (LACRI) of the Brazilian Agricultural Research Company (EMBRAPA) in the municipality of Sete Lagoas, Minas Gerais State, Brazil.

Artificial diets with honey and water (Table 1) were used in laboratory tests and the other ingredients adjusted to obtain a pasty texture due to the hygroscopic potential of the honey combined with water. This facilitates its placement with a spatula at the sides of rearing containers, and feeding of the predator. The diet was based on pet food (Table 1) that has been used for more than a year in the LACRI to rear *Doru luteipes* (Scudder) (Dermaptera: Forficulidae). This diet without water has a powdery consistency and is offered to the predator at the bottom of the rearing units.

Adults of *E. connexa* fed on unfrozen eggs of *A. kuehniella* and on artificial diet 2 (Table 1) were mated to obtain 40 larvae per treatment. One day after hatching, individual larvae were placed inside 50 ml plastic vials covered with transparent acrylic and fixed on a piece of Styrofoam.

Table 1 Composition of the artificial diets supplied to *Eriopis connexa* at 25±1°C, 12 h photophase and 70±10% r.h.; all data are given in grams

Compound (g)	Artificial diet 1	Artificial diet 2	Artificial diet 3	Artificial diet 4
Honey	100	100	100	–
Brewer's yeast	100	100	90	46
Soybean crumbs	–	–	5	–
Wheat germ	–	–	5	54
FeSO ₄	–	1.5	1.5	–
Ascorbic acid	1.5	1.5	1.5	–
Propionic acid	0.5	0.5	0.5	–
Sorbic acid	0.25	0.25	0.25	–
Nipagin	0.25	0.25	0.25	0.25
Milk	–	–	–	28
Cat food ^a	–	–	–	72
Water	60	60	60	–

^a Corn, flour of gut, meat and bones; rice; soybean crumbs; fish flour; animal fat; gluten; bran; vegetable oil, mineral vitamin premix; lysine; potassium chloride; DL-methionine; sodium chloride and taurine

The diets were supplied *ad libitum* to *E. connexa* larvae according to the treatment (Table 2) until emergence of adults. These adults were sexed and weighed just after emergence and transferred to glass rearing cages (12 cm diam and 18 cm height) covered with PVC film and fed according to the treatment.

The larvae and adults of *E. connexa* were observed daily to evaluate the number of instars ($n=20$); duration of each instar, periods of prepupa and pupa stages and from larva to adult ($n=20$); viability of larva, prepupa and pupa and from larva to adult ($n=40$); besides weight and sex ratio of this predator ($n=40$).

The experimental design was completely randomized with 17 treatments (diets; Table 2) with four replications, each one with ten *E. connexa* larvae. A completely randomized block design was used. All the larvae used in the experiments were from a laboratory colony fed on frozen eggs of *A. kuehniella* and artificial diet 2 (Table 1).

Data were submitted to analysis of variance (ANOVA) and the averages compared by the Tukey test at 5% probability through the computer program MSTAT-C, version 2.1 (Michigan State University).

Results

Eriopsis connexa had four instars (Table 3). Most larvae of this predator which were fed with only

artificial diet died in the first instar. The larval stage of *E. connexa* fed on *A. kuehniella* eggs frozen for 6 months, alone or in combination with artificial diets, was of longer duration (Table 3); the duration of the larval stage varied between treatments. The longest duration of the larval stage of *E. connexa* with eggs of *A. kuehniella* (frozen for 6 months) separately or in combination with artificial diets (Table 3), may be due to food type (Table 3). In treatments T13 and T17, the first instar of *E. connexa* was of shorter duration than the other instars (Table 3).

The viability of the larval stage of *E. connexa* was higher with eggs of *A. kuehniella* frozen for 1 day, and fed separately or in combination with artificial diets. On the other hand, treatment T9 (eggs of *A. kuehniella* frozen for 6 months) and the artificial diet 1 (Table 5) had lower values. The development of *E. connexa* with artificial diet 1 in combination with eggs of *A. kuehniella* frozen for 6 months or 1 day (T9 and T14, respectively) was higher than with the other diets tested. Besides, the duration of the larval stage of this predator was longer and the viability lower for individuals fed with eggs of *A. kuehniella* only (6 months' freezing), or in combination with artificial diets (Table 5), except in treatment T9, which had 60% adult emergence.

The duration of the prepupal stage varied from 1.0 to 1.4 days (Table 4), without any difference between treatments. The viability of *E. connexa* prepupae was

Table 2 Diet supplied to *Eriopsis connexa* larvae at $25\pm 1^\circ\text{C}$, 12 h photophase and $70\pm 10\%$ r.h. in the 17 different treatments

Treatment no.	Food
T1	Artificial diet 1
T2	Artificial diet 2
T3	Artificial diet 3
T4	Artificial diet 4
T5	Artificial diet 1 + artificial diet 4
T6	Artificial diet 2 + artificial diet 4
T7	Artificial diet 3 + artificial diet 4
T8	Eggs of <i>A. kuehniella</i> (frozen for 6 months)
T9	Eggs of <i>A. kuehniella</i> (frozen for 6 months) + artificial diet 1
T10	Eggs of <i>A. kuehniella</i> (frozen for 6 months) + artificial diet 2
T11	Eggs of <i>A. kuehniella</i> (frozen for 6 months) + artificial diet 3
T12	Eggs of <i>A. kuehniella</i> (frozen for 6 months) + artificial diet 4
T13	Eggs of <i>A. kuehniella</i> (frozen for 1 day)
T14	Eggs of <i>A. kuehniella</i> (frozen for 1 day) + artificial diet 1
T15	Eggs of <i>A. kuehniella</i> (frozen for 1 day) + artificial diet 2
T16	Eggs of <i>A. kuehniella</i> (frozen for 1 day) + artificial diet 3
T17	Eggs of <i>A. kuehniella</i> (frozen for 1 day) + artificial diet 4

Table 3 Mean duration (days; at 25±1°C, 12 h photophase and 70±10% r.h.) of each instar of *Eriopsis connexa* receiving ten different diets

Treatment no.	Instars			
	First	Second	Third	Fourth
T8	3.9±0.2 ab ^a	4.2±0.1 a	2.9±0.3 ab	5.9±0.3 a
T9	3.4±0.1 abc	2.1±0.1 b	2.9±0.2 ab	5.2±0.3 abc
T10	3.5±0.2 abc	2.6±0.1 b	3.0±0.2 ab	4.3±0.3 bcd
T11	3.9±0.1 ab	2.7±0.2 b	2.9±0.2 ab	5.8±0.7 a
T12	4.0±0.2 a	3.1±0.1 ab	3.8±0.1 a	5.6±0.1 ab
T13	3.3±0.1 bc	2.9±0.2 ab	2.7±0.2 b	3.6±0.1 d
T14	3.1±0.1 a	2.9±0.1 ab	2.9±0.1 ab	3.9±0.2 cd
T15	3.0±0.2 a	2.7±0.2 b	3.3±0.1 ab	4.2±0.1 cd
T16	3.1±0.1 a	2.3±0.1 b	3.3±0.02 ab	4.0±0.2 cd
T17	3.0±0.1 a	2.8±0.1 ab	3.2±0.3 ab	4.2±0.3 cd
CV (%)	7.2	21.2	12.6	12.1
ANOVA	($F=10.6799$, $df=27$, $P<0.2084$)	($F=3.4073$, $df=27$, $P<0.4430$)	($F=2.7563$, $df=27$, $P<0.2375$)	($F=9.6182$, $df=27$, $P<0.0026$)

^a Within columns, means followed by a common letter do not differ by the Tukey test ($P<0.05$)

similar on all diets, except for individuals fed only with *A. kuehniella* eggs frozen for 6 months.

The diet used affected the duration of the pupal stage of *E. connexa* with values between 3.7 and 4.9 days (Table 4) and with viability from 78.6% to 100.0% (Table 5).

The duration from larva to adult of *E. connexa* varied from 17.1 to 23.1 days between treatments (Table 4). The longer and shorter duration from larva to adult was found with *A. kuehniella* eggs frozen for 6 months (T8) or 1 day (T13), respectively (Table 4). The production of *E. connexa* adults was higher when its larvae were fed on *A. kuehniella* eggs frozen for 1 day (T13) with 92.5% viability or for 1 day

associated with the artificial diets 1 or 2 (T14 and T15), with 100.0% and 92.5% (Table 5), respectively. The duration from larva to adult of *E. connexa* was 17.1, 17.9 and 18.3 days with diets T13, T14 and T15, respectively (Table 5).

The sex ratio of *E. connexa* was similar between treatments, from 0.19 to 0.58 (Table 6). In general, males are lighter than females, but the body weight of *E. connexa* differed between treatments. The lighter and heavier females and males of this predator weighed 6.2 and 3.9 mg with eggs of *A. kuehniella* eggs frozen for 6 months and 12.3 and 8.4 mg, respectively, with eggs frozen for 1 day and given in combination with artificial diet 1 (Table 6).

Table 4 Duration (days; at 25±1°C, 12 h photophase and 70±10% r.h.) of the larval, prepupal and pupal stages, and from larva to adult of *Eriopsis connexa* receiving ten different diets

Treatment no.	Larvae	Prepupae	Pupae	Larva–adult
T8	16.9±0.8 a ^a	1.4±0.1 a	4.9±0.1 a	23.2±0.9 a
T9	13.6±0.6 bc	1.0±0.0 a	4.1±0.1 bc	18.7±0.6 bcd
T10	13.4±0.2 bc	1.0±0.0 a	4.0±0.00 c	18.4±0.2 bcd
T11	15.3±0.6 ab	1.4±0.3 a	4.7±0.2 ab	21.4±0.7 abc
T12	16.5±0.4 a	1.1±0.1 a	4.1±0.1 bc	21.7±0.4 ab
T13	12.4±0.4 c	1.0±0.0 a	3.7±0.01 c	17.1±0.4 d
T14	12.8±0.1 c	1.0±0.0 a	4.1±0.1 bc	17.9±0.1 cd
T15	13.2±0.2 bc	1.0±0.1 a	4.1±0.1 bc	18.3±0.2 bcd
T16	12.7±0.2 c	1.2±0.1 a	3.9±0.2 c	17.8±0.2 d
T17	13.2±0.3 bc	1.0±0.0 a	4.2±0.2 bc	18.4±0.4 bcd
CV (%)	6.6	20.2	6.3	5.0
ANOVA	($F=11.8194$, $df=27$, $P<0.0303$)	($F=2.2217$, $df=27$, $P<0.1849$)	($F=7.5040$, $df=27$, $P<0.2970$)	($F=16.6308$, $df=27$, $P<0.0569$)

^a Within columns, means followed by a common letter do not differ by the Tukey test ($P<0.05$)

Table 5 Viability (%; at $25\pm 1^\circ\text{C}$, 12 h photophase and $70\pm 10\%$ r.h.) of the larval, prepupal and pupal stages, and from larva to adult of *Eriopis connexa* receiving ten different diets

Treatment	Larvae	Prepupae	Pupae	Larva–adult
T8	42.5±4.8 bc ^a	82.3±2.8 b	78.6±4.3 c	27.5±7.5 de
T9	72.5±10.3 ab	100.0±0.0 a	82.7±2.2 bc	60.0±5.8 bcd
T10	45.0±8.7 bc	93.7±6.2 a	94.1±5.9 abc	40.0±7.1 cde
T11	42.5±7.5 bc	100.0±0.0 a	88.2±7.0 abc	37.5±7.5 de
T12	22.5±4.8 c	100.0±0.0 a	100.0±0.0 a	22.5±4.8 e
T13	92.5±4.8 a	100.0±0.0 a	100.0±0.0 a	92.5±4.8 ab
T14	100.0±0.0 a	100.0±0.0 a	100.0±0.0 a	100.0±0.0 a
T15	92.5±2.5 a	100.0±0.0 a	100.0±0.0 a	92.5±2.5 ab
T16	82.5±10.3 a	100.0±0.0 a	100.0±0.0 a	82.5±10.3 ab
T17	75.0±8.7 ab	100.0±0.0 a	96.7±2.3 a	72.5±10.3 abc
CV (%)	20.9	4.4	7.1	21.3
ANOVA	($F=14.1767$, $df=27$, $P<0.0009$)	($F=7.0817$, $df=27$, $P<0.0032$)	($F=5.9031$, $df=27$, $P<0.2243$)	($F=19.1007$, $df=27$, $P<0.0000$)

^a Within columns, means followed by a common letter do not differ by the Tukey test ($P<0.05$)

Discussion

The number of instars of *E. connexa* was the same as reported for *Coccinella undecimpunctata* (L.) (Coleoptera: Coccinellidae) fed on *Megoura persicae* (Buckton) and *Aphis fabae* (Scopoli) (Hemiptera: Aphididae; Cabral et al. 2006), and for this predator fed on prey *D. noxia* and *R. maidis*, *D. noxia*, *A. pisum* and *M. persicae*, *Cinara atlantica* (Wilson) (Hemiptera: Aphididae; Miller 1995; Miller and Paustian 1992; Oliveira et al. 2004); and for *Scymnus* (*Neopullus*) *sinuanodulus* Yu and Yao (Coleoptera: Coccinellidae) with foliage of *Tsuga canadensis* (L.) infested with Adelgidae (Lu et al. 2002). Inadequate foods can increase the number of instars, which

shows that those supplied to *E. connexa* had similar nutritional quality because this predator presented the same number of instars in the treatments in which adults were obtained, as reported for *Stenomacrus catenifer* Walsingham (Lepidoptera: Elachistidae) with natural and artificial diets (Navas and Parra 2005).

The artificial diet used alone did not allow the development of larvae of *E. connexa*, but it affected the pre-imaginal parameters of this predator. This suggests the need for additional nutrients to artificial diets such as essential amino acids and mineral salts due to the generalist feeding behavior of Coccinellidae, which can feed on different prey and on pollen grains (Berkvens et al. 2008; Iperiti 1999; Isikber and Copland 2002; Ragkou et al. 2004). Coccinellidae rarely com-

Table 6 Sex ratio and adult weight (mg) of *Eriopis connexa* obtained from larvae fed on ten different diets at $25\pm 1^\circ\text{C}$, 12 h photophase and $70\pm 10\%$ r.h.

Treatment no.	Sex ratio	Weight (mg)	
		Females	Males
T8	0.37±0.05 b ^a	6.2±0.5 d	3.9±0.2 e
T9	0.39±0.09 b	10.2±1.0 abc	4.6±0.3 de
T10	0.58±0.08 ab	9.5±0.3 bc	5.4±0.2 cd
T11	0.37±0.04 b	8.1±0.7 cd	4.5±0.2 de
T12	0.78±0.07 a	7.6±0.3 cd	3.7±0.4 e
T13	0.48±0.05 ab	11.0±0.5 ab	7.0±0.3 a
T14	0.55±0.06 ab	12.3±0.3 a	8.4±0.1 a
T15	0.57±0.09 a	11.2±0.2 ab	8.2±0.4 a
T16	0.52±0.09 ab	11.0±0.3 ab	7.6±0.3 ab
T17	0.52±0.05 ab	9.1±0.3 bc	6.2±0.3 bc
CV (%)	28.9	11.2	10.4
ANOVA	($F=2.7533$, $df=27$, $P<0.3850$)	($F=12.2060$, $df=27$, $P<0.0000$)	($F=4.8683$, $df=27$, $P<0.0002$)

^a Within columns, means followed by a common letter do not differ by the Tukey test ($P<0.05$)

pleted the first instar when supplied with inadequate food. However, poor food quality can be associated with a specific nutrient that limits the development of the predator (Michaud 2005). *Coleomegilla maculata fuscilabris* (Mulsant) (Coleoptera: Coccinellidae) did not complete its development when supplied with only *Toxoptera citricida* (Kirkaldy) (Hemiptera: Aphididae) but this predator survived with this prey when given in combination with an appropriate artificial diet during the first instar (Michaud 2000).

The duration of the larval stage of *E. connexa* fed on *A. kuehniella* eggs frozen for 1 day, separately or together with artificial diets, was similar (Table 3) to the 3.1, 2.2, 2.5 and 3.0 days, respectively for the first, second, third and fourth instars for this predator fed on *C. atlantica* (Navas and Parra 2005), but with higher values than those with *D. noxia* and *R. padi*: 8.1 days (Michaud 2005), *C. atlantica*, 10.82 days (Navas and Parra 2005), and *D. noxia*, *A. pisum* and *M. persicae*, 6.3–7.2, 8.1–8.4, 9.7–8.5 days, respectively (Michaud 2000).

Eggs of *A. kuehniella* frozen for 6 months were not adequate for *E. connexa* development. The increase in the duration of the larval stage is common for insects receiving poor nutrient diets and may be due to their nutritional demands (Scriber and Slansky 1981; Stathas 2000; Thompson 1999). Food quality was found to affect the development and survival of Coccinellidae larva (Isikber and Copland 2002; Kalushkov and Hodek 2001, 2004). This shows the necessity of further studies because frozen eggs can be stored to rear predatory insects with reduced costs compared to fresh prey. On the other hand, the freezing period of *A. kuehniella* eggs affected its nutritive quality (Mohaghegh and Amir-Maafi 2007).

The longest duration of the fourth instar in all treatments may be due to the need of *E. connexa* larvae to obtain nutrients for pupation and metamorphosis as found for other insects (Scriber and Slansky 1981; Stathas 2000).

The artificial diets in combination with eggs of *A. kuehniella* reduced the duration of the larva stage of *E. connexa* compared with individuals fed only with eggs of *A. kuehniella* frozen for 6 months. The use of one food type was inadequate for *E. connexa*, which had better results with artificial diet 1 in combination with eggs of *A. kuehniella* independent of the freezing period. This may be explained by the generalist behavior of predatory Coccinellidae that feed on

different prey and pollen (Berkvens et al. 2008; Ipert 1999; Isikber and Copland 2002; Ragkou et al. 2004).

The duration of the prepupal stage without any difference between treatments had similar values to those of *H. convergens* with eggs of *A. kuehniella* (1516); *C. sanguinea* fed on *M. persicae*, *Megoura viciae* (Buckton); *Aphis gossypii* (Glover) or *A. fabae* (Isikber and Copland 2002) and *Stethorus punctillum* (Weise) (Coleoptera: Coccinellidae) with *Tetranychus urticae* (Koch) (Acari: Tetranychidae; Ragkou et al. 2004).

The prepupal stage of *E. connexa* starts when the larva stops feeding and remains attached with its last abdominal segment to the surface of the cover or to the lateral parts of the cage. The prepupae of this predator may exhibit abrupt movements when touched, such as observed for *Olla v-nigrum* (Mulsant) (Coleoptera: Coccinellidae; Kato et al. 1999a). This indicates that the food received during the larval stage does not affect the prepupae of this predator. However, the viability of this stage for individuals fed only with *A. kuehniella* eggs frozen for 6 months was shorter than those of the other treatments (Table 5). This suggests that the artificial diets associated with eggs improved the development of *E. connexa*, and that the predator transfers reserves stored between instars to the adult stage.

The prepupae of *E. connexa* exhibited a different behavior from that of *S. sinvanodulus*, which stopped feeding, released a great amount of liquid from the anal area, and became immobile for 1–2 days (Lu et al. 2002). This was observed also for *E. connexa*, but some larvae of *S. sinvanodulus* exhibited different behavior (Lu et al. 2002) from that of *E. connexa*: they started crawling after the immobility period and molted to pupae after the crawling period. However, a high percentage of *S. sinvanodulus* pupae that exhibited this behavior died (Lu and Montgomery 2001; Lu et al. 2002). Crawling of *S. sinvanodulus* was associated with a mechanism for appropriate feeding used by their larvae to disperse to protected places for pupation (Lu et al. 2002).

The reduction of the duration of the pupal stage is important for biological control programs, to obtain adults quickly to increase populations of natural enemies (Auad 2003; Nordlund and Greenberg 1994). The pupa viability of *E. connexa* had similar values to those of this predator with *C. atlantica* (Oliveira et al. 2004) or *D. noxia* and *R. padi* (Miller

1995; Miller and Paustian 1992). The duration of the pupal stage of *E. connexa* was short, but with high viability in treatments T13 and T16.

The sex ratio (number of males/ number of males + females) of *E. connexa* had similar values to those of this predator fed on *M. persicae*, *D. noxia* or *A. pisum* (Michaud 2005) and *C. atlantica* (Nordlund and Greenberg 1994) and to the coccinellid predators *H. convergens*, *Cryptolaemus montrouzieri* (Mulsant), *Delphastus pusillus* (LeConte), *Harmonia axyridis* (Pallas), *Lindorus lophanthae* (Blais) and *Stethorus punctillum* (Weise) (Heimpel and Lundgren 2000).

Heavier females are common for Coccinellidae (Iperti 1999; Kato et al. 1999a; Sighinolfi et al. 2008). In all treatments *E. connexa* females were heavier than males, suggesting that females can be differentiated from males based on body weight, as reported for *Hippodamia convergens* (Guérin-Méneville) and *Coccinella septempunctata* (L.) (Coleoptera: Coccinellidae) fed on *Myzus persicae nicotianae* (Sulzer) (Hemiptera: Aphididae; Katsarou et al. 2005).

The greater weight of *E. connexa* females may be due to their reproduction, which is a physiological process regulated by nutrients stored in the female body (Wheeler 1996; Zanuncio et al. 2002). Generally, larger females are more fertile than smaller ones. An example is seen in *Cycloneda sanguinea* (L.) (Coleoptera: Coccinellidae), which is heavier and has higher fecundity when fed on *Toxoptera citricida* (Kirkaldy) compared with *Aphis spiraecola* (Patch) (Hemiptera: Aphididae) (Michaud 2000).

Eggs of *A. kuehniella* frozen for 1 day and fed alone (T13) or in combination with artificial diet 1 (T14) are more appropriate for mass rearing *E. connexa*, resulting in shorter duration of immature stages and heavier adults of this predator. This is important because the weight indicates the amount of nutrients stored, which can affect mating, dispersion, flight and fecundity in insects (Nordlund and Greenberg 1994; Omkar et al. 2006). On the other hand, the longer duration of immature stages and the lower weight of *E. connexa* fed on eggs of *A. kuehniella* frozen for 6 months suggest the need for additional nutrients in this diet in order to rear this predator. The artificial diets, separately, are inadequate for *E. connexa* to complete its development, but their results can be better when given along with *A. kuehniella* eggs. The association of artificial diet 1 with eggs of *A.*

kuehniella represents the best diet for rearing *E. connexa*.

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