Artemia cysts as an alternative food for the predatory bug
Macrolophus pygmaeus

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Abstract

The suitability of cysts of the brine shrimp Artemia sp. as a factitious food for the predator Macrolophus pygmaeus Rambur was investigated. The influence of decapsulation time and hydration of the cysts on the performance of the predator were studied in the absence of plant material. A longer time of decapsulation had a positive influence on the development of the predator. Hydration of cysts had a significant impact on nymphal survival when cysts were non-decapsulated or poorly decapsulated. An experiment in which nymphs were switched from a diet of hydrated cysts to non-hydrated cysts showed that in the absence of plant material the relative importance of hydrating the cysts decreased with nymphal age. Especially, the first instar and to a lesser extent the second instar appear to be susceptible to water shortage. Effects of prolonged rearing on development and reproduction on brine shrimp cysts from different origins were tested in the presence of plant material. Rearing M. pygmaeus on Artemia sp. (Jingyu Lake) cysts yielded similar survival, development, adult weight and fecundity in the fourth as in the second generation. In contrast, for Artemia franciscana cysts, an increase in nymphal development was notable. Biochemical analyses showed that total amino acid content and the concentration of the different amino acids did not differ among diets and generations. There were, however, differences in total fatty acid content between the different diets and generations and in the concentration of certain fatty acids, indicating that insects fed brine shrimp cysts may show nutritional deficiencies compared to those reared on a diet of Ephesia kuehniella eggs. Our results indicate that decapsulated brine shrimp cysts are an economically viable alternative food source in at least part of the rearing process for M. pygmaeus.

Introduction

Macrolophus pygmaeus Rambur is a polyphagous predator that is successfully used as a biological control agent against several crop pests in European greenhouses (Perdikis and Lykouressis 2000; Martínez-Cascles et al. 2006). One of its main drawbacks is its relatively high rearing cost, mainly due to the intensive use of eggs of the Mediterranean flour moth Ephesia kuehniella Zeller as a food source. The market price of these eggs is currently around 500–600 EUR/kg. Alternative, cheaper food sources may reduce the production costs and thus stimulate the use of this mirid in biological control (De Clercq 2004). The brine shrimp Artemia spp. can be such an alternative. These branchiopod crustaceans are routinely used as food sources in aquaculture (Lavens and Sorgeloos 1996) and their cysts are an
order of magnitude cheaper than E. kuehniella eggs (Arijs and De Clercq 2001). Cysts of Artemia sp. were successfully tested as a food for the anthocorid predator Orius laevigatus (Fieber) (Arijs and De Clercq 2001 and De Clercq et al. 2005). Further, Callebaut et al. (2004), Riuadavets et al. (2006) and Castañe et al. (2006) also reported that the mirid predator Macrolophus caliginosus Wagner can successfully develop and reproduce on a diet of Artemia cysts.

In above studies, the tested predators were offered decapsulated or non-decapsulated cysts that were or were not hydrated [by hydrating the cysts in water, their water content increases up to 90% (De Clercq et al. 2005)]. However, no attention was given to the degree to which the cysts were decapsulated. Decapsulation is the process of removing the chorion that encysts the Artemia embryo; this can be made by short-term exposure to a concentrated hypochlorite solution (Lavens and Sorgeloos 1996). First, this study investigated the effect of extent of decapsulation on food value for M. pygmaeus. Second, the importance of hydrating brine shrimp cysts when supplied for use as a food during nymphal development was evaluated. In these experiments, no plant materials were offered throughout nymphal development to avoid masking effects of plant feeding. Finally, the developmental effects of prolonged rearing of M. pygmaeus on Artemia cysts from different geographic origins and with different physical and biochemical characteristics were assessed. Carcass analyses were performed to compare predators reared on Artemia cysts with conspecifics reared on E. kuehniella eggs in terms of amino acid and fatty acid composition.

**Materials and Methods**

**Insect colony**

Individuals of M. pygmaeus were obtained from a laboratory colony started with eggs supplied by Koppert B.V. (Berkel en Rodenrijs, The Netherlands). The nymphs were reared in plastic containers (24 × 16 × 8 cm), furnished with paper towels and covered with nylon netting. They were reared on leaves of Nicotiana tabacum L. cv. Xanthi and were fed frozen eggs of E. kuehniella, also supplied by Koppert B.V.: the eggs were kept in the freezer for a maximum of 1 month prior to being fed to the predators. E. kuehniella eggs were replenished every 2 days. A paper plug soaked with tap water fitted into a plastic dish (4.3 cm diameter) was used as a water source. The containers were maintained in growth chambers at 23 ± 1°C, 60 ± 5% RH and a 16L : 8D h photoperiod.

**Effect of decapsulation time of Artemia franciscana cysts on development and fecundity of M. pygmaeus in the absence of plant material**

Cysts of Artemia franciscana (San Francisco Bay, USA) were obtained from the Laboratory of Aquaculture & Artemia Reference Center of Ghent University. An initial experiment was carried out to test the effect of decapsulation time on the value of brine shrimp cysts as a food for M. pygmaeus; this was performed for both dry and hydrated cysts of A. franciscana. The hydration and decapsulation processes for Artemia cysts have been described by Lavens and Sorgeloos (1996). The standard decapsulation procedure for A. franciscana cysts consisted of hydration of the cysts, followed by subjecting them to a decapsulation process of 150–180 s, at which time the colour of the cysts turned from brown to orange. At that moment, the chorion is fully dissolved whereas the hatching membrane and embryo are not affected by the hypochlorite solution. To test the effect of exposure time to the hypochlorite solution on the quality of the cysts, the duration of the decapsulation process was varied, with a shorter time (85 ± 5 s) leading to a lower degree of decapsulation (as observed under a stereomicroscope) and a longer time (245 ± 5 s) after which not only the chorion is dissolved but also the hatching membrane is affected. No plant material was used in this experiment given that Callebaut et al. (2004) showed that plant material has a masking effect on nymphal performances of M. caliginosus on different diets.

First instars (<24 h old) were taken out of the stock cultures and were placed individually in plastic cups (4 cm diameter, 2.5 cm high) on wax paper substrates (absorbent household paper drenched in beeswax). A parafilm capsule filled with tap water was offered to the insects as an alternative moisture source, according to the methods described by Vandekerkhove et al. (2006). Predator nymphs were subjected to one of five treatments: feeding on E. kuehniella eggs; dry decapsulated cysts, with the decapsulation procedure stopped at 85 s; dry decapsulated cysts, with the decapsulation procedure stopped at 245 s; hydrated decapsulated cysts, with the decapsulation procedure stopped at 85 s; hydrated decapsulated cysts, with the decapsulation procedure stopped at 245 s. The foods were replenished on Mondays, Wednesdays and Fridays and old foods were discarded weekly to avoid fungal growth in the containers. The water capsules were replaced after
when needed. Nymphs that died on the first day of the experiment were replaced by new ones, as it was assumed that their death was probably due to handling rather than to treatment effects. Nymphal developmental time and survival were recorded on a daily basis. Nymphs that successfully reached the adult stage were sexed and weighed at emergence on a Sartorius Genius balance type ‘ME215P’ with 0.01 mg precision (Sartorius, Goettingen, Germany). Adults were then paired and placed on a fresh tobacco leaf disc; they were given the same diet as during their nymphal life. After 1 week, female adults were dissected and oocytes were counted following the method described by Vandekerkhove et al. (2006): late vitellogenic to mature oocytes were scored 1, early to mid vitellogenic oocytes 0.5 and previtellogenic oocytes 0.25, mature oocytes present in the oviducts were also scored 1. The scores for all ovarioles were then summed yielding a weighted sum of oocytes. At the same time, the leaf discs were examined for oviposited eggs as described by Ferran et al. (1996). The containers were maintained in growth chambers at $23 \pm 1^\circ C$, $60 \pm 5\%$ RH and a $16L:8D$ h photoperiod.

**Effect of hydration of A. franciscana cysts on nymphal development of M. pygmaeus in the absence of plant material**

To evaluate the importance of hydrating cysts before presenting them to the predator nymphs, an experiment was set up in which nymphs were switched at different ages from feeding on hydrated non-decapsulated A. franciscana (San Francisco Bay) cysts to feeding on dry non-decapsulated cysts. Like in the above experiment, no plant material was supplied to the insects. First instars ($<24$ h old) taken from the stock cultures were placed individually in plastic cups (4 cm diameter, 2.5 cm high) on wax paper substrates and offered a parafilm capsule with tap water.

Predators were subjected to one of the following treatments: dry cysts supplied from the first nymphal instar up to dissection of adults (DN1); switching to dry cysts from the second nymphal instar on (DN2); switching to dry cysts from the third nymphal instar on (DN3); switching to dry cysts from the fourth nymphal instar on (DN4); switching to dry cysts from the fifth nymphal instar on (DN5); and switching to dry cysts from the adult stage on (DAd). Food and water were supplied as described above. Nymphal developmental time and survival were recorded daily and resulting adults were weighed at emergence. Adults were paired and females were dissected after 1 week to count oocytes using the methods described above. The experiment was carried out in a growth chamber at $23 \pm 1^\circ C$, $60 \pm 5\%$ RH and a $16L:8D$ h photoperiod.

**Prolonged rearing of M. pygmaeus on Artemia cysts from different origins**

To assess the value of Artemia cysts for prolonged rearing of M. pygmaeus, the predator was exclusively reared on this food during four successive generations. In this experiment, the predator was reared either on *E. kuehniella* eggs (control), on *A. franciscana* cysts from San Francisco Bay, USA, or on *Artemia* cysts originating from Jingyu Lake, Xinjiang, China (ARC code 1524). The latter cysts belonged to a further non-determined *Artemia* species, but were selected because of their characteristics similar to *A. tibetiana*, i.e. higher content in highly unsaturated fatty acids (HUFA) and their larger size than *A. franciscana* (San Francisco Bay) cysts (Van Stappen et al. 2003). All cysts were decapsulated for 150 s and were offered in non-hydrated form. First instars ($<24$ h old) were taken out of the stock culture and were placed in Plexiglas cylindrical containers (9 cm diameter, 3.5 cm high) provided with ventilation openings screened with fine mesh. In each container, a small sweet pepper plant (*Capsicum annuum* L., var. California Wonder) was placed, with its roots immersed in water. Approximately 30 first instars from the stock culture were transferred to each of nine containers; these nymphs were allowed to develop to adulthood on either of the tested foods. This was called generation 1 (G1). The foods were placed on the bottom of the cylinders and replenished on Mondays, Wednesdays and Fridays. When these nymphs had developed into adults, the sweet pepper plant was taken out of the cylinder and replaced by a new one every 3 days. The removed plant was then transferred to a new cylinder to allow deposited eggs to hatch. To assess developmental and reproductive performance, about 30 newly hatched nymphs (G2) were collected from the rearing cylinders and placed individually on tobacco leaf discs. The isolated nymphs were offered their respective diet and developmental parameters were recorded. Nymphs that successfully reached the adult stage were sexed, weighed and paired. Adult pairs were placed on a fresh tobacco leaf disc and were given the same diet as during their nymphal stages. After 1 week, females were dissected and oocytes were counted; the leaf discs were examined for oviposited eggs as described above.
The remaining nymphs were maintained in the communal rearing cylinders and reared up to a fourth generation of nymphs (G4). Then again, ca. 30 nymphs were isolated to determine biological parameters as was performed in G2. In addition, newly emerged females of G2 and G4 were subjected to biochemical carcass analyses. Amino acid analysis and lipid analysis were performed according to the methods described by Zapata et al. (2005).

**Statistical analysis**

A fully randomized experimental design was used. A one-way analysis of variance (ANOVA) was performed to determine differences in nymphal development times, adult weights and fecundity data among treatments. Mean differences were distinguished by using Duncan’s test or Tamhane’s T2 test for unequal variances. Mean values were significantly different when P < 0.05. All data were statistically analysed using spss 15.0 (SPSS Inc. 1989–2007).

**Results**

**Effect of decapsulation time of A. franciscana cysts on development and fecundity of M. pygmaeus in the absence of plant material**

Nymphal survival of *M. pygmaeus* was the highest when given *E. kuehniella* eggs (85.7%) (table 1).

**Table 1** Nymphal developmental time and survival, adult weight, oocyte counts and number of eggs laid per female in the first week of adult life of *Macrolophus pygmaeus* fed on eggs of *Ephestia kuehniella* or on dry or hydrated cysts of *Artemia franciscana* exposed to decapsulation for 85 or 245 s

<table>
<thead>
<tr>
<th>Diet</th>
<th>Developmental time (days)</th>
<th>Nymphal survival, % (n)</th>
<th>Adult weight (mg)</th>
<th>Weighted sum of oocytes (n)</th>
<th>No. eggs laid (n)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ephestia kuehniella</em></td>
<td>17.0 ± 0.2 a (13)</td>
<td>17.5 ± 0.4 a (11)</td>
<td>85.7 (28)</td>
<td>0.81 ± 0.02 a (13)</td>
<td>1.32 ± 0.03 a (11)</td>
</tr>
<tr>
<td><em>Artemia</em> dry decapsulated for 85 s</td>
<td>24.0 ± 0.4 c (4)</td>
<td>24.2 ± 1.3 c (5)</td>
<td>32.1 (28)</td>
<td>0.58 ± 0.01 c (4)</td>
<td>0.87 ± 0.02 c (5)</td>
</tr>
<tr>
<td><em>Artemia</em> dry decapsulated for 245 s</td>
<td>18.6 ± 0.4 b (14)</td>
<td>20.2 ± 1.0 b (6)</td>
<td>71.4 (28)</td>
<td>0.70 ± 0.02 b (14)</td>
<td>1.09 ± 0.03 b (6)</td>
</tr>
<tr>
<td><em>Artemia</em> hydrated decapsulated for 85 s</td>
<td>18.4 ± 0.6 b (9)</td>
<td>20.0 ± 0.4 b (12)</td>
<td>75.0 (28)</td>
<td>0.74 ± 0.03 ab (9)</td>
<td>1.05 ± 0.02 b (12)</td>
</tr>
<tr>
<td><em>Artemia</em> hydrated decapsulated for 245 s</td>
<td>18.8 ± 0.3 b (12)</td>
<td>19.4 ± 0.6 ab (9)</td>
<td>75.0 (28)</td>
<td>0.72 ± 0.03 b (12)</td>
<td>1.09 ± 0.04 b (9)</td>
</tr>
</tbody>
</table>

Mean values (±SE) within a column followed by the same letter are not significantly different (P > 0.05; Duncan’s test).
females fed hydrated cysts exposed longer to decapsulation was significantly lower than that in the control (F = 3.5; d.f. = 4.36; P < 0.01; ANOVA), despite yielding a high weighted sum of oocytes. This is in contrast with earlier findings showing a good correlation between weighted sum of oocytes and number of eggs laid (Vandekerkhove et al. 2006).

Effect of hydration of *A. franciscana* cysts on nymphal development of *M. pygmaeus* in the absence of plant material

Figure 1 demonstrates the relative importance of offering hydrated cysts for the survival of the nymphal predators. 83.3% of the nymphs that only received dry cysts (DN1) never reached adulthood, with a mortality percentage during the first two nymphal stadia reaching 60%. Mortality decreased as nymphs were allowed to feed longer on hydrated cysts before being switched to dry cysts. Mortality was the lowest (20%) for nymphs that were fed exclusively on hydrated cysts.

Table 2 shows that developmental time generally increased as the nymphs were switched earlier in life from hydrated to dry cysts (males: F = 11.4; d.f. = 5.55; P < 0.001; females: F = 3.9; d.f. = 5.38; P = 0.006; ANOVA). When dry cysts were offered from the first or the second nymphal instar on, this resulted in significantly longer development of males than when offered dry cysts from the third instar on. Developmental time of females was significantly shorter when fed on hydrated cysts from the first to fifth instar (DAd), to the fourth instar (DN4) or to the third instar (DN4), than when offered dry cysts from the first instar.

Weights of adult males (F = 6.7; d.f. = 5.55; P < 0.001; ANOVA) and females (F = 9.9; d.f. = 5.38; P < 0.001; ANOVA) tended to decrease as they were switched to dry cysts earlier in nymphal development. Adult males weighed significantly less when they were solely fed on dry cysts or on hydrated cysts up to the fourth nymphal instar as compared with males that were exclusively fed on hydrated cysts or switched to dry cysts from the fifth nymphal instar on. Females had superior body weights when they were always offered hydrated cysts.

Only females fed exclusively dry cysts as nymphs had significantly lower oocyte counts than those who received hydrated cysts at least from the third instar on (F = 4.9; d.f. = 5.36; P = 0.002; ANOVA).

Prolonged rearing of *M. pygmaeus* on *Artemia* cysts from different origins

Table 3 shows the biological parameters for *M. pygmaeus* reared during successive generations on different diets. Nymphal survival was high on all diets varying from 84.8% to 100% in G2 and from 90.0% to 93.7% in G4. Whereas in G2 there was no difference in developmental time between *E. kuehniella* eggs and *A. franciscana* cysts, nymphs took longer to develop on both *Artemia* cyst diets than on *E. kuehniella* eggs in G4 (males: F = 22.3; d.f. = 5.94; P < 0.001; females: F = 12.1; d.f. = 5.67; P < 0.001; ANOVA). Males and females took longer to develop on *A. franciscana* cysts in G4 than in G2. Developmental times

![Fig. 1 Mortality in consecutive nymphal instars (N1–N5) of *Macrolophus pygmaeus* when fed exclusively on non-decapsulated dry cysts (DN1) or non-decapsulated hydrated cysts (DAd) of *Artemia franciscana*, or when switched from non-decapsulated hydrated to non-decapsulated dry cysts from the second (DN2), third (DN3), fourth (DN4), or fifth (DN5) instar on.](image-url)
of predator nymphs on *Artemia* sp. (Jingyu Lake) cysts were, however, similar in G2 and G4. Male weights were not affected by treatment (F = 0.62; d.f. = 5.94; P = 0.69; ANOVA). For females, adult weights showed only differences in the second generation, with the control diet yielding higher body weights than either *Artemia* cyst diet (F = 4.3; d.f. = 5.67; P = 0.002; ANOVA). The number of eggs oviposited in the first week of adult life did not differ among treatments (F = 1.0; d.f. = 5.59; P = 0.42; ANOVA), nor did oocyte counts upon dissection (F = 2.1; d.f. = 5.60; P = 0.78; ANOVA), although the *A. franciscana* cysts resulted in somewhat lower oocyte counts and oviposition compared to the other diets in both generations. There were no significant differences in development and reproductive performances in the same generation of the predator on either *Artemia* cyst type.

Contents of fatty acids and amino acids determined by carcass analysis of females reared on different diets in G2 and G4 are reported in table 4. Total fatty acid content was significantly higher for predators fed *E. kuehniella* eggs than on the *Artemia* diets (F = 14.9; d.f. = 4.16; P < 0.001; ANOVA). Compared to the diet of *E. kuehniella* eggs, insects fed brine shrimp cysts showed deficiencies in oleic (C18:1), linoleic (C18:2), and, to a lesser extent, palmitic acid (C16:0) in both generations tested (fig. 2).

Table 4 shows that the total amino acid content of *M. pygmaeus* females did not differ among diets and
generations (F = 0.13; d.f. = 4,19; P = 0.97; ANOVA). The values for each amino acid (fig. 3) also showed no differences between the different diets and generations. The main amino acids found in the carcasses of nymphs on the different diets were (expressed as % of total amino acid content) aspartate (8.3–8.5%), glutamate (9.8–10.4%), glycine (8.6–8.9%), alanine (10.1–10.8%) and to a lesser extent leucine (7.6–7.9%), valine (6.5–6.6%), serine (6.0–6.2%), lysine (5.8–6.5%) and proline (5.8–6.1%).

**Table 4** Mean total fatty acids and total amino acids in carcasses of *Macrolophus pygmaeus* fed with eggs of *Ephestia kuehniella*, or with cysts of *Artemia franciscana* or *Artemia* sp. (Jingyu Lake) during consecutive generations

<table>
<thead>
<tr>
<th>Diet/ generation</th>
<th>Total fatty acids, µg/mg fresh weight (n)</th>
<th>Total amino acids, nmol/mg fresh weight (n)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ephestia</em> kuehniella/G2</td>
<td>85.1 ± 2.6 a (5)</td>
<td>1146 ± 29 a (5)</td>
</tr>
<tr>
<td><em>Artemia</em> franciscana/G2</td>
<td>53.2 ± 3.1 c (4)</td>
<td>1128 ± 33 a (5)</td>
</tr>
<tr>
<td><em>Jingyu</em> Artemia/G2</td>
<td>53.5 ± 7.2 c (3)</td>
<td>1111 ± 97 a (5)</td>
</tr>
<tr>
<td><em>Artemia</em> franciscana/G4</td>
<td>67.3 ± 3.0 b (5)</td>
<td>1144 ± 112 a (5)</td>
</tr>
<tr>
<td><em>Jingyu</em> Artemia/G4</td>
<td>58.11 ± 3.0 bc (4)</td>
<td>1132 ± 90 a (5)</td>
</tr>
</tbody>
</table>

G2, generation 2; G4, generation 4. Means (±SE) within a column followed by the same letter are not significantly different (P > 0.05; Duncan’s test).

**Discussion**

When testing the effect of decapsulation time on the value of brine shrimp cysts as a food for *M. pygmaeus*, survival was similar to that reported by Castanè et al. (2006), who offered dry or hydrated non-decapsulated *Artemia* cysts to *M. caliginosus* nymphs provided with *Sedum rubrotinctum* R.T. Clausen (Crassulaceae) leaf cuttings as plant material. Mahdian et al. (2006) found that hydrated decapsulated *A. franciscana* cysts were an inadequate food for the predatory stinkbugs *Podisus maculiventris* (Say) and *Picromerus bidens* (L.). Arijs and De Clercq (2001) reported that it was not possible to rear *O. laevigatus* on dry cysts, as nymphs of this predator failed to reach the second instar on this food. They concluded that cysts had to be hydrated for successful development of this predator. Castanè et al. (2006) recorded a similar development for *M. caliginosus* given *E. kuehniella* eggs (ca. 18 days) as in our study for *M. pygmaeus*, even though in their study the experiment was carried out at 25°C and the insects were provided with plant material. For *M. caliginosus* nymphs fed brine shrimp cysts, latter authors recorded developmental times ranging from 17.5 to 19.5 days. Comparison of the results is complicated by the fact that different species were tested at different climatic regimes and in the presence or absence of plant material.

From our study, it appears that the longer the decapsulation time of the *Artemia* cysts, the better the developmental performance of the nymphs is.

**Fig. 2** Fatty acids in carcasses from females of *Macrolophus pygmaeus* reared on eggs of *Ephestia kuehniella*, cysts of *Artemia franciscana* or cysts of *Artemia* sp. (Jingyu Lake) during consecutive generations; G2: generation 2, G4: generation 4; within fatty acids, bars with the same letter are not significantly different among treatments (P > 0.05; Duncan’s test).
Besides complete removal of the cyst shell, longer decapsulation times may also cause damage to the hatching membrane, and as such make the embryo more easily available for feeding predators. When the cysts are hydrated, they may be easier to penetrate by the predator's stylets; in that case, the decapsulation process is probably of less importance. Thus, exposing non-hydrated cysts for a longer time to the decapsulation treatment makes them a more suitable food for M. pygmaeus. Non-hydrated cysts should be preferred as they lead to fewer problems with moulds in the rearing containers than fully hydrated cysts, which contain more than 90% water (De Clercq et al. 2005). Preliminary studies indicated that in cultures where plant materials are supplied, dry cysts (ca. 4% water) hydrate when they are on a leaf surface until they reach a water content of 20% or more within 24 h, facilitating feeding by the bugs.

Hydrated cysts resulted in better nymphal development than non-hydrated cysts when they were poorly decapsulated. Hydration of poorly or non-decapsulated cysts may make them easier to penetrate by the predators. When the cysts are completely decapsulated, hydration is of little importance. The longer the predators had access to non-decapsulated hydrated cysts in their nymphal life, the better their chances of survival were. The relative importance of offering hydrated A. franciscana cysts decreased with nymphal age: access to hydrated cysts proved of greatest importance for the first two instars. Small nymphs of M. pygmaeus have the most difficulty piercing the outer cyst shell with their rostrum. Likewise Arijs and De Clercq (2001) reported that O. laevigatus also had difficulties penetrating the hard outer alveolar layer of dry non-decapsulated cysts, which led them to conclude that for this predator hydration of the cysts was necessary for successful development.

Thompson (1999) pointed out that insects may show a loss in fitness only after they have been reared on a diet for multiple generations, as nutritional deficiencies may only then become expressed. Coudron et al. (2002) noted that insects may also adapt to a new diet, resulting in a gradual improvement of biological parameters. In this study, even after four consecutive generations of rearing on brine shrimp cysts, there was no significant decrease in survival, adult weight and fecundity of the predator. However, on A. franciscana cysts, immature development took significantly longer in the fourth generation than in the second, whereas this was not the case on Artemia sp. (Jingyu Lake) cysts, suggesting that latter cysts are nutritionally superior to the former. This may be related to the higher content in HUFA, or alternatively to the larger size of Artemia sp. (Jingyu Lake) cysts (ca. 320 μm) compared to A. franciscana cysts from San Francisco Bay (ca. 227 μm) (Van Stappen et al. 2003; Camargo et al. 2005). The predator may have needed to spend less time and energy to handle such larger cysts and extract nutrients from them. Castaño et al. (2006) did not find differences in any of the discussed biological parameters of

**Fig. 3** Total amino acids in carcasses of Macrolophus pygmaeus females reared on eggs of Ephestia kuehniella, cysts of Artemia franciscana or cysts of Artemia sp. (Jingyu Lake) during consecutive generations; G2: generation 2, G4: generation 4; mean values for each amino acid were not significantly different among treatments (P > 0.05; Duncan’s test).
M. caliginosus offered different strains of Artemia sp. (species was not identified in the study), neither in the first, nor in the sixth generation; their experiment included a strain with low HUFA content and one with high HUFA content.

Besides data on developmental and reproductive performance, biochemical analysis may allow a further evaluation of the quality of insects reared on alternative foods (Grenier and De Clercq 2003). Amino acid contents of carcasses of M. pygmaeus females did not differ between those fed E. kuehniella eggs and those fed Artemia cysts, even after four generations of rearing. Fatty acid contents of M. pygmaeus females reared on Artemia cysts showed deficiencies in palmitic, oleic and linoleic acid as compared to those reared on flour moth eggs, indicating that these fatty acids were either unavailable for the predators or present at insufficient levels in the cysts. Most of the fatty acids consumed by insects are stored as energy sources in neutral lipids, and some others have a key role as structural elements incorporated in membrane phospholipids or as signal molecules (Downer 1985; Stanley 2000).

It is well accepted that specific lipids are necessary dietary requirements for all insects, particularly the immature stages (Reinecke 1985). Many insects require dietary polyunsaturated fatty acids to support growth and reproduction, and either linoleic or linolenic acid usually satisfies their nutritional need (Canavoso et al. 2001). Development on A. franciscana cysts is slower in G4 as compared to G2, but this is not reflected in the biochemical carcass analysis of the insects. This ‘generation effect’ only occurred for insects fed A. franciscana cysts but not for those fed Artemia sp. (Jingyu Lake) cysts. Interestingly, the amino acid and fatty acid patterns of M. pygmaeus adults correspond well to those reported for eggs of E. kuehniella (De Clercq et al. 2005).

In conclusion, non-hydrated and fully decapsulated cysts of Artemia sp. proved to be a viable alternative food source for rearing M. pygmaeus, provided that plant materials were offered. Although hydrated cysts also constitute a good food, their use is not recommended as they are more prone to microbial contamination, especially when fully decapsulated. However, some loss of fitness was observed after several generations of exclusive rearing on certain types of Artemia materials. Artemia cysts may nonetheless constitute an economically viable alternative food for the production of M. pygmaeus, as it may reduce inputs of expensive lepidopteran eggs in parts of the production process of this important biocontrol agent.

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References


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