Phytoparasitica (2017) 45:175–182
DOI 10.1007/s12600-017-0581-2

Investigation on the storage possibilities of *Rhyzobius lophantae* Blaisdell (Coleoptera: Coccinellidae) at different temperatures and periods

D. Şenal · O. Demirözer · İ. Karaca

Received: 17 August 2016 / Accepted: 16 April 2017 / Published online: 1 May 2017

Abstract Storage of natural enemies at low temperatures has an important role in biological control programs. It is an very critical factor that the stored individuals is used without losing their characters such as longevity, survival, fecundity, etc. In this study, the storage possibilities of *Rhyzobius lophantae* Blaisdell (Coleoptera: Coccinellidae) adults under low temperatures (4 °C and 12 °C) for four different periods (10, 20, 30 and 40 days) were investigated. The survival rates and life span of adults (male and female), daily and total egg numbers laid by females stored at 4 °C and 12 °C for 10, 20, 30 and 40 days were determined. The survival rates of adults stored at 4 °C for 10 and 20 days were found to be 94.54% and 58.02% respectively. However, no adults stored at 4 °C for 30 and 40 days were observed to survive. The highest survival rate was 88% for the adults stored at 12 °C for 10 days. The average daily laid egg numbers were 12.51 and 9.37, and total egg numbers were 850.30 and 738.30 of adults stored at 4 °C for 10 and 20 days, respectively. The decrease in daily and total egg numbers of females stored at 12 °C was observed with increasing storage period. There was no significant difference in longevities of male and female stored at 4 °C and 12 °C. These results show that short-term storage at low temperatures does not affect certain properties of the predator when compared to 12 °C and storage periods.

Keywords Storage possibilities · Biological control · Coccinellidae · *Rhyzobius lophantae*

Introduction

Rhyzobius lophantae Blaisdell (Coleoptera: Coccinellidae) is an important natural enemy of most armored scales of Diaspididae family (Flores and Carlson 2009; Stathas 2000a; Stathas 2000b; Stathas et al. 2002; Uygun 1981) and it has the characteristics of undiapaused, high mobility, well prey selectivity, adult longevity, high fecundity, rapid development (five to seven generation in a year) and lack of parasitism (Flores and Carlson 2009; Stathas 2000b). These superior abilities have allowed the effectively use of *R. lophantae* in biological control of armored scale insects in the USA, Italy, Argentina, Bermuda, Algeria, Tunisia, Morocco, Georgia, Greece, Texas, Pacific (Cave et al. 2013; Flores and Carlson 2009; Stathas 2000b). The recent studies pointed out that, it has become fortuitously established on Cycad Aulacaspis Scale, *Aulacaspis yasumatsui* (Hemiptera: Diaspididae) in South Texas, USA since October, 2006 (Flores and Carlson 2009). Furthermore, the researchers mentioned that cycad aulacaspis scale was controlled by *R. lophantae* and *Aphytis lignanensis* after established. On the other hand, *R. lophantae* was introduced to Guam (2004), Rota (2007) and Palau (2009) in the Pacific, and released on native

D. Şenal (✉)
Faculty of Agricultural and Natural Sciences, Department of Plant Protection, Bilecik Şeyh Edebali University, Bilecik, Turkey
e-mail: derya.senal@bilecik.edu.tr

O. Demirözer · İ. Karaca
Faculty of Agriculture, Department of Plant Protection, Süleyman Demirel University, Isparta, Turkey

cycads infested by *A. yasumatsui*. The predator rapidly established and controlled heavily infested plants (Anonymous 2010; Cave et al. 2013).

The practices used in biological control are realized as the mass production and periodic releases of natural enemies or adaptation and import of new beneficial species, in such cases the beneficial populations in the agroecosystem do not adequately control the pests (Uygun 1991; Uygun et al. 2010). Sufficiently large numbers of beneficials in periodic releases or shipped of some natural enemies in cooled containers are increasing the importance of storage at low temperatures in biological control (Ghazy et al. 2014; Luczynski et al. 2008). Moreover, developing storage techniques are very important to store insects when the colony is not in use, to assist in accumulating sufficient numbers of insects for innundative releases, or to reduce costs of retaining a colony between inoculative releases (Abdel-Salam and Abdel-Baky 2000; Chang-Chun et al. 2012; Coudron et al. 2007; Gagne and Coderre 2001; Hofsvang and Hagvar 1977; Jalali and Singh 1992; Özgökçe et al. 2006; Tauber and Tauber 1978; Tauber et al. 1993). Thus, storage of natural enemies may permit economic and limited production, stockpiling during low production or unexpected periods of high demand, rearing to occur at a time of the year (Riddick and Wu 2010). In order to be able to use natural enemies effectively in biological control practices, a certain number of storages are required under appropriate circumstances without adversely affecting the basic features of the beneficial such as reproductive potential, development, life span, consumption, survival, etc. (Ghazy et al. 2014; Özgökçe et al. 2006; Riddick and Wu 2010). For all that reasons, the beneficials have to be kept in perfect storage conditions and techniques.

It is known that low temperatures slow down insect's metabolism and activities (Colinet et al. 2006; Ladurantaye et al. 2010). Furthermore, prolonged exposure to low temperatures could cause cellular injury, thermal shock, direct-chilling injury, spontaneous nucleation of body water, metabolism perturbations or accumulation of toxins and could be fatal for the insects (Chen et al. 1987; Gagne and Coderre 2001). Storage periods and low temperatures are vital importance for the without losing their biological and physiological characteristics. Furthermore the storage at appropriate conditions is advantageous in delaying activity and preventing cannibalism of beneficials (Ladurantaye et al. 2010).

There are several studies related with storage of coccinellids at low temperatures. *Adalia bipunctata* L. (Hamalanien 1977), *Adonia variegata* (Borodkin 1973), *Chilocorus bipustulatus* L. (Şenal et al. 2001), *Coccinella septempunctata* L., *C. undecimpunctata* L. (Abdel-Salam and Abdel-Baky 2000), *Coleomegilla maculata lengi* Timberlake (Gagne and Coderre 2001), *Cryptolaemus montrouzieri* Mulsant (Özgökçe et al. 2006; Yiğit et al. 1994), *Harmonia axyridis* (Abdel-Salam et al. 1997; Chang-Chun et al. 2012; Deng 1982), *Nephus includens* Kirsch (Yiğit et al. 1994) and *Rodolia cardinalis* (Quezada and DeBach 1973). These studies have investigated that the cold storage affected some biological characteristics of ladybirds. *C. undecimpunctata* adults may survive extended periods of storage at 6 °C compared to the other developmental stages. Larval stage of predator didn't survived after 30 and 60 days (Abdel-Salam and Abdel-Baky 2000). Özgökçe et al. (2006) found that the longevity, total fecundity and daily reproduction of *C. montrouzieri* adults kept at 15 °C for 5 days were 69.7, 686.7 and 9.8, respectively. However, these values decreased in prolonged storage periods. For the first time, the storage possibilities of *R. lophanthae* at storage periods and different low temperatures was investigated by the present study.

Determination of the storage conditions for predators should direct us to establish suitable low temperature and periods for long-term storage, transport, contributing to release. In this study it was aimed to determine the post storage effects and storage potential of *R. lophantae* adults at different low temperatures for certain periods were evaluated by measuring survival, male and female longevities, reproductive capacity. Additionally, in order to determine performance of stored individuals, statistical analaysis was performed on survival rate, lifespan of adults and egg numbers.

Material and methods

Mass rearing of *Aspidiotus nerii* Bouché

The Oleander Scale, *Aspidiotus nerii* Bouché (Hemiptera: Diaspididae) was used as prey to feed *R. lophantae.* The crawlers of *A. nerii* obtained from infested shoots and leaves of *Acacia cyanophylla* Lindley, (Fabaceae) at Çukurova University Campus, Adana, Turkey and transfered to potato tubers. The oleander scale reared on potato tubers. As the individuals on potatoes continue to reproduce, new potato tubers were placed on a weekly basis to ensure continuity of rearing. Mass rearing of *A. nerii* were realized in the chamber room at 26 ± 1 °C, 60% relative humidity and 16:8 (L:D) photoperiod.

Mass rearing of *Rhyzobius lophantae* Blaisdell

The adults of *R. lophantae* were collected from Cyprus acacia trees infested by *A. nerii* at Çukurova University Campus. The adults transferred to plexiglass boxes (width 13 cm, length 25 cm) including potato tubers infested by *A. nerii.* Prey for *R. lophantae* added regularly according to the prey shortage in the rearing boxes. The plexiglass boxes were kept in the chamber room at 26 ± 1 °C, 60% relative humidity and 16:8 (L:D) photoperiod.

Storage studies of *Rhyzobius lophantae* Blaisdell at low temperatures for different periods

The experimental design was a randomized complete plot and ten replications were made for each trial. In the study, eleven adults of *R. lophantae* newly emerged from pupae were transferred to each ventilated plastic petri dishes (6 cm in diameter) for each temperature and storage period. 110 adults of *R. lophantae* were used in each trial. The petri dishes were initially placed at 4 and 12 °C and were kept separetely in the incubators for 10, 20, 30 and 40 days under full darkness and 60% relative humidity. The adults were taken to the climate chambers at 16:8 (L:D) photoperiod, 26 ± 1 °C and 60% RH after completing the storage periods at 4 and 12 °C and kept for 24 h. Afterwords, dead or alive adults were counted at each replication. The dead individuals were removed and coded at each replications. Alive adults were kept at climatic chambers to use for further experiments.

The potato pieces (about 2–3 cm^2) infested by *A. nerii* were placed in the petri dishes for feeding of individuals, and this potato pieces were replaced daily with new ones in order to determine the egg numbers of *R. lophantae*. The number of eggs laid by *R. lophantae* was counted every 24 h under stereo-microscope (40X zoom) during the experiment.

There were few individuals crushed or escaped in replications by accident and the data of these individuals didn't evaluated in the statistical analysis for life span of female-male and fecundity.

The preparations of dead adults

The preparation of dead adults coded during the experiment was made according to Uygun (1981) to determine total-daily laid egg numbers of females, and genders in the petri dishes. The samples were kept for one day with diluted phenol. After softening, the abdomens were separated from the bodies of the predator using a pin and then transferred to a 10% KOH solution. Between 12 and 24 h, they were immersed in distilled water. The male and female were determined by removing their genital organs.

Statistical analysis

Data on the survival rate, longevity, fecundity of *R. lophantae* adults for storage periods (10, 20, 30 and 40 days) at each temperature (4 °C and 12 °C) were subject to two-factor analysis of variance. The comparisons of means for each treatment were made with Tukey HSD (Honestly Significant Difference) test ($p \leq 0.05$). The Statistical software package SPSS 18.0 was used for all statistical analyses.

Results

Effects of different storage temperatures and periods on the survival rates of adults

The data demonstrates that the survival rates showed a rapid decrease according to storage duration and all adults died at 4 °C for 30 and 40 days storage periods. Although the highest survival rate was observed as 94.54% at the end of the storage period of 10d at 4 °C, the survival rates of adults stored at 12 °C was higher than 4 °C (Fig. 1). Durations of storage at both temperatures significantly affected the survival rates of stored adults (F = 25.5792, df = 1–78, $P \leq 0.01$). The adult survival rates varied significantly among storage periods of 10, 20, 30 days at 4 °C (F = 192.4841, df = 3–36, P≤,0001). At the same time, there were statistically significant differences between survival rates of the adults stored at 12 °C for 10 and 40 storage days (F = 5.4135, df = 3–36, $P \leq 0.0035$).

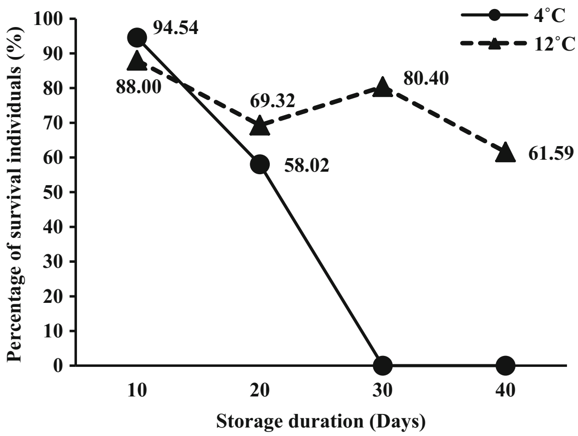

Fig. 1 Survival rates (%) of *Rhyzobius lophantae* adults stored at 4 and 12 °C for 10, 20, 30 and 40 days

Effect of different storage temperatures and periods on life span of *Rhyzobius lophantae* female and male

Adults were held at 26 ± 1 °C, 16:8 (L:D) photoperiod and 60% RH at post-storage period to determine the impacts of storage on adult life span. All adults died during storage periods of 30 and 40 days at 4 °C. Male and female longevities varied according to storage periods at each temperature. Males (78.31 days) stored at 4 °C for 10 days lived longer than the females (69.80 days), but females (91.27 days) survived longer than males (62.25 days) stored at the same temperature for period of 20 days. It was found that male longevity (65.76 days) of *R. lophantae* after stored at 12 °C for 10 days was longer than females (53.02 days). In contrast, females at other storage periods (20, 30 and 40 days) survived longer than males (Fig. 2, Table 1).

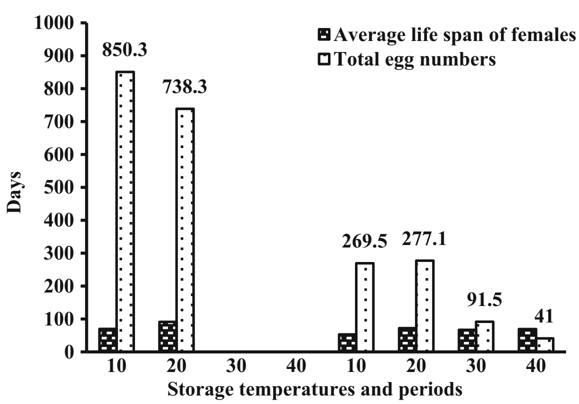

Fig. 2 The total egg numbers and life span of *Rhyzobius lophantae* female following four different storage periods at 4 and 12 °C

However, the results of statistical analaysis indicate that storage periods for each temperature had no significant effects on life span. (F = 37.9044, df = 3–36, P≤,0001 for 4 °C, F = 2.1426, df = 3–36, $p \leq 0.1119$ for 12 °C in female; F = 15.0328, df = 3–36, P≤,0001 for 4 °C, F = 0.1864, df = 3–36, $p \leq 0.905$ for 12 °C in male) (Table 1).

The longest lifespan of females and males were 91.27 days after storage period of 20 days and 78.31 days for storage period of 10 days at 4 °C. The differences in both female and male longevities at 4 °C and 12 °C for each storage period (10, 20, 30 and 40 days) were significant (F = 9.5976, df = 1–78, $P \leq 0.0027$ for female; F = 10.6052, df = 1–78, $P \leq 0.0017$ for male).

Surviving curves showed a gradual decrease in the lifespan of adult stage after storage periods at different temperatures except storage period of 20 days at 4 °C in Fig. 3. It was observed a sharp decline (drop) in the curve for storage period of 20 days at 4 °C. The adult mortality occured at the beginning of the cohort. The earliest mortality occurred 112nd days in the adults stored at 12 °C for 40 days amongst all storage periods (Fig. 3).

Effect of different storage temperatures and periods on fecundity of *Rhyzobius lophantae* female

The data in Table 1 and Fig. 2 illustrates that females stored at 4 °C laid higher number of eggs than those at 12 °C. The differences between two temperatures for total egg numbers were found statistically significant (F = 8.8827, df = 1–78, $P \leq 0.0038$). In contrast, there was not a significant differences between two temperatures for daily egg numbers (F = 3.3895, df = 1–78, $P \leq 0.0694$). The females laid no eggs at 4 °C because adults died at the storage periods of 30 and 40 days. The maximum daily and total egg numbers throughout the life span of females stored at 4 °C for 10 days were 12.51 and 850.30 respectively (Table 1).

Females stored at 12 °C for 10–20 days and 30–40 days laid similar daily and total of egg numbers. No significant differences in daily egg numbers were found between the storage periods of 10 and 20 days, and between the storage periods of 30 and 40 days. On the other hand, significant differences in daily and total egg numbers between storage periods of 10–20 days and 30–40 days were found (F = 17.8588, df = 3–36, P≤,0001 for daily egg numbers and F = 17.4945, df = 3–

Table 1 The life span (day) of female-male and daily-total egg numbers of *Rhyzobius lophantae* after storage at two different temperatures and four different periods (Mean ± S.E)

Temp. (°C)	Storage Period (day)	Female n	Female Life span		Male n	Male Life span		Daily Egg		Total Egg	
4	10	46	69.80 ± 5.02	a	58	78.31 ± 6.8	a	12.51 ± 1.11	a	850.30 ± 63.81	a
	20	29	91.27 ± 14.53	a	36	62.25 ± 20.08	a	9.37 ± 1.67	a	738.30 ± 133.19	a
	30	0	0	b	0	0	b	0	b	0	b
	40	0	0	b	0	0	b	0	b	0	b
12	10	47	53.02 ± 4.20	a	42	65.76 ± 6.40	a	6.42 ± 0.56	a	269.50 ± 18.63	a
	20	35	72.04 ± 4.72	a	41	58.16 ± 8.82	a	4.69 ± 0.97	a	277.10 ± 52.22	a
	30	54	67.01 ± 5.07	a	35	63.03 ± 5.71	a	1.91 ± 0.52	b	91.50 ± 15.16	b
	40	31	69.59 ± 8.34	a	27	65.00 ± 9.96	a	0.62 ± 0.09	b	41.00 ± 7.90	b

n total number of surviving individuals

*Means followed by the same letters in a column at each temperature are not significantly different (Tukey, $p \leq 0.05$)

36, P≤,0001 for total egg numbers). Additionally, egg numbers decreased rapidly with increasing storage periods (Table 1 and Fig. 2). The fecundities of females were affected by storage periods and low temperatures.

In Fig. 3, fecundity curves was almost static in all storage periods. Reproductive peak was reached at the beginning of the oviposition period. Reproduction stored at 12 °C for 30 and 40 days demonstrated no conspicuous peaks. High fecundity values reached at the beginning of the adult stage, but they decreased substantially toward the end of lifespan except for adults stored at 12 °C for 30 and 40 days (Fig. 3).

Discussion and conclusion

The studies have been conducted to determine the effects of storage at low temperatures for many biological control agents on survival, fecundity, life span, egg viability, consumption capacity, etc. in different developmental stages. The studies indicated that their biological characteristics were influenced by species, biotypes, preconditioning, developmental stage, storage temperature and storage period (Abdel-Salam et al. 1997; Abdel-Salam and Abdel-Baky 2000; Chang-Chun et al. 2012; Colinet et al. 2006; Deng 1982; Gagne and Coderre 2001; Hofsvang and Hagvar 1977; Jalali and Singh 1992; Özgökçe et al. 2006; Şenal et al. 2001; Tauber et al. 1993).

The results of this study show that low temperatures and storage periods affect the survival of *R. lophantae* adults. The survival rates of adults were more than 50% in different storage periods at 12 °C. Survival decreased rapidly and adults died at storage periods of 30 and 40 days at 4 °C. The death of adults can be related with the cellular damage, metabolism perturbations or accumulation of toxins at low temperatures and storage periods (Gagne and Coderre 2001). Yiğit et al. (1994) studied on storage possibilities of *Cryptolaemus montrouzieri* Mulsant and *Nephus includens* Kirsch. *C. montrouzieri* lived maximum 10 days at 7 °C and 22 days at 15 °C. On the other hand, 50% survival rate of *N. includens* adult populations obtained for 8.27 days and 11.26 days at 7 °C and 15 °C respectively. *N. includens* lived maximum 20 days at 7 °C and 25 days at 15 °C. Chang-Chun et al. (2012) found that *Harmonia axyridis* (Pallas) survived more than 80% when they were stored for 150 days at 3 °C and 6 °C. Abdel-Salam and Abdel-Baky (2000) studied developmental stages of *Coccinella undecimpunctata* L. were stored at 6 °C for 7, 15, 30, 45 and 60 days in a refrigerator. The survival rates of adults newly emerged from pupae were 25, 15 and 10% for storage periods of 7, 15 and 30 days, but no adults survived for storage periods of 45 and 60 days. The mortality rates of *C. montrouzieri* were increased with increasing periods at 15 °C, especially those longer than 5 days (Özgökçe et al. 2006). The differences between the results can be attributed to the different predator species.

In addition to survival, life span of male and female at 4 °C and 12 °C was also determined in this study. There was no differences in life span of females and males stored at each temperature for each storage periods. Abdel-Salam and Abdel-Baky (2000)

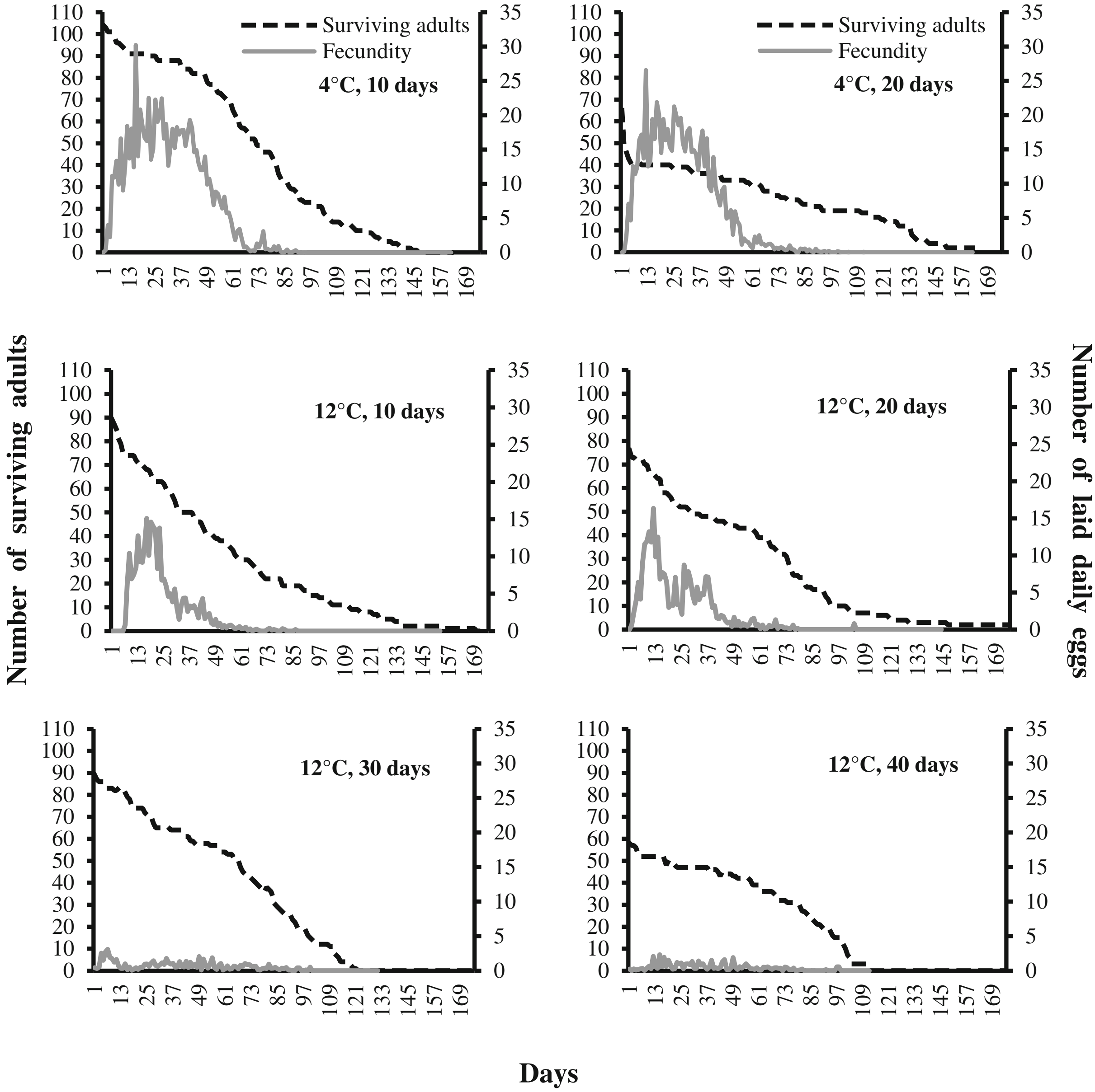

Fig. 3 Surviving and fecundity curves of *Rhyzobius lophantae* adult following different storage periods (10, 20, 30 and 40 days) at two temperatures (4 and 12 °C)

determined that male and female longevities of *Coccinella undecimpunctata* were respectively 35 and 42.3 days for 7 storage period at 6 °C. Male and female of predator stored for 15 days at 6 °C lived respectively 31 and 40 days. Results showed that storage period had a significant impact on adult longevity. Longevities of *Cryptolaemus montrouzieri* female after storage periods (5, 10, 15, 20 days) at 15 °C were respectively 69.7, 57.3, 73.4 and 56.8 days. Adult longevity of predator was also affected by cooling periods (Özgökçe et al. 2006).

Compared to the individuals that develop without being stored and the stored individuals of *R. lophanae*, male and female longevities were respectively 43.1 and 53.2 days at 25 °C (Nar et al. 2009). In the other studies, adult longevity of *R. lophantae* was 121.3 days at 25 °C (Stathas 2000a). When the results are compared with studies of Nar et al. (2009) and Stathas (2000a), the life spans of stored adults are longer than those of unstored individuals. This can be attributed to slow-down in insect metabolism and activity because of storage at low temperatures (Colinet et al. 2006; Ladurantaye et al. 2010). As a result of these studies, adult longevities are affected by different storage periods and temperatures.

Chang-Chun et al. (2012) found that long-term cold storage had different effects on the fecundity of *H. axyridis* at different temperatures. The adults stored 90-days at 0 °C had the largest reproductive capacity. The female stored 90 days at 0 °C, 90–150 days at 3 °C and 120–150 days at 6 °C laid similar egg numbers. Egg numbers increased up to 120 day storage at 6 °C and decreased at −3 °C. Özgökçe et al. (2006) reported that total fecundities of *C. montrouzieri* after storage periods of 5, 10, 15, 20 days at 15 °C were 686.7, 478.5, 366.4 and 385.8 respectively; while daily reproductions were also 9.8, 6.4, 3.0 and 3.1 respectively. Nar et al. (2009) determined that the daily and total egg numbers of *R. lophantae* were 13.6 and 680 respectively during oviposition period at 25 °C. Stathas (2000b) found that female of *R. lophantae* laid mean 633.7 eggs during oviposition period at 25 °C and 65% R.H. In both studies, similar results obtained in the egg numbers. In this storage study, it is pointed that the egg numbers laid by surviving female stored at 4 °C for 10 and 20 storage period were similar with results of Nar et al. (2009) and Stathas (2000b). But, the egg numbers decreased with prolonged storage periods at both temperatures.

As a result of this study, we found that low temperatures for each storage periods affected mortality, fecundity and longevity of *R. lophanae* adults. While life spans of male and female were not affected, egg numbers were influenced by storage periods for each temperatures. Survival and fecundities of *R. lophantae* stored at 4 °C and four different periods decreases according to increasing storage periods. Long storage periods had a direct impact on the decrease of the number of eggs. This study suggests that *R. lophantae* can be stored at low temperatures for short periods without any loss of biological characteristics. *R. lophantae* should be used in short periods during the cross-country transport.

Future studies should focus on the feeding and performance (predation on pests) of stored predators in the field.

Acknowledgements Authors thank to Prof. Dr. M. Bora Kaydan for contribution to the article. This study was financially supported by Scientific Research Projects of University of Çukurova as the Project ZF. 2000. 17.

References

Abdel-Salam, A. H., & Abdel-Baky, N. F. (2000). Possible storage of *Coccinella undecimpunctata* (Col., Coccinellidae) under low temperature and its effect on some biological characteristics. *J. Appl. Ent., 124*, 169–176.

Abdel-Salam, A. H., Ellington, J. J., El-Adl, M. A., Abou El-Naga, A. M., & Ghanim, A. A. (1997). Effect of cold storage on survival, longevity and fecundity of *Harmonia axyridis* adults. *Proceedings of the 1st Nat., Conf.*, Vol. 1, Mansoura, Egypt (pp. 58–72).

Anonymous. (2010). *Insect activity-Cycad aulacaspis scale (CAS) in Micronesia. Forest health 2009 highlights. Pacific Islands* (pp. 6–7).

Borodkin, I. (1973). Naturschutz und biologischer Pflanzenschutz. *Intern. Z. Landw., 4*, 394–397.

Cave, R. D., Chao, C.-T., Kumashiro, B. R., Marler, T. E., Miles, J., Moore, A., Muniappan, R., & Watson, G. W. (2013). Status and biological control of cycad Aulacaspis Scale. *Biocontrol News and Information, 34*(1), 1N–8N.

Chang-Chun, R., Wen-Mei, D., Xiu-Mei, W., Jun-Jie, Z., & Lian-Sheng, Z. (2012). Effect of long-term cold storage on the fitness of pre-wintering *Harmonia axyridis* (Pallas). *BioControl, 57*, 95–102.

Chen, C. P., Delinger, D. L., & Lee, R. E. (1987). Cold-shock injury and cold rapid hardening in the flesh fly, *Sarcophaga crassipalpis*. *Physiological Zoology, 60*, 297–304.

Colinet, H., Renault, D., Hance, T., & Vernon, P. (2006). The impact of fluctuating thermal regimes on the survival of a cold-exposed parasitic wasp, *Aphidius colemani*. *Phsiological Entomology, 31*, 234–240.

Coudron, T. A., Ellersieck, M. R., & Shelby, K. S. (2007). Influence of diet on long-term cold storage of the predator *Podisus maculiventrsi* (Say) (Heteroptera: Pentatomidae). *Biological Control, 42*, 186–195.

Deng, D. A. (1982). Experiments on feeding with artificial diets and cold storage of *Harmonia axyridis* Pallas. *Insects Knowledge, 19*, 11–12.

Flores, D., & Carlson, J. (2009). Fortiotus establishment of *Rhyzobius lophantae* (Coleoptera: Coccinellidae) and *Aphytis lignanensis* (Hymenoptera: Encyrtidae) in South Texas on the cycad Aulacaspis Scale, *Aulacaspis yasumatui* (Hemiptera: Diaspididae). *Southwestern Entomologist Scientific Note, 34*(4), 488–492.

Gagne, I., & Coderre, D. (2001). Cold storage of *Coleomegilla maculata* larvae. *Biocontrol Science and Technology, 11*, 361–369.

Ghazy, N. A., Suzuki, T., Amano, H., & Ohyama, K. (2014). Air temperature optimisation for humudity-controlled cold storage of the predatory mites *Neoseiulus californicus* and *Phytoseiulus persimilis* (Acari: Phytoseiidae). *Pest Management Science, 70*, 483–487.

Hamalanien, M. (1977). Storing dormant *Coccinella septempunctata* and *Adalia bipunctata* (Col., Coccinellidae) adults in the laboratory. *Ann. Agric. Fenn., 16*, 184–187.

Hofsvang, T., & Hagvar, E. B. (1977). Cold storage tolerance and supercooling points of mummies of *Ephedrus cerasicola* Stary and *Aphidius colemani* Viereck (Hym. Aphidiidae). *Norwegian Journal of Entomology, 24*(1), 1–6.

Jalali, S. K., & Singh, S. P. (1992). Differential response of four *Trichogramma* species to low temperatures for sort term storage. *Entomophaga, 37*, 159–165.

Ladurantaye, Y., Khelifi, M., Cloutier, C., & Coudron, T. A. (2010). Short-term sorage conditions for transport and farm delivery of the stink bug *Perillus bioculatus* for the biological control of the Colorado potato beetle. *Canadian Biosystems Engineering, 52*, 1–7.

Luczynski, A., Nyrop, J. P., & Shi, A. (2008). Pattern of female reproductive age classes in mass-reared populations of *Phytoseiulus persimilis* (Acari: Phytoseiidae) and its influence on population characteristics and quality of predators following cold storage. *Biological Control, 47*, 159–166.

Nar, E., Ulusoy, M. R., & Karaca, İ. (2009). The influence of different temperatures on the development of *Rhyzobius lophantae* Blaisdell (Coleoptera: Coccinellidae). *SDÜ Fen Bilimleri Dergisi, 4*(1), 59–74.

Özgökçe, M. S., Atlihan, R., & Karaca, İ. (2006). The life table of *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae) after different storage periods. *Journal of Food, Agriculture and Environment, 4*(1), 282–287.

Quezada, J. R., & DeBach, P. (1973). Bioecological and population studies of cottony-cushion scale, *Icerya purchasi* Mask. and its natural enemies, *Rodolia cardinalis* Mul. and *Cryptochaetum iceryae* Will. in Southern California. *Hilgardia, 41*, 631–688.

Riddick, E. W., & Wu, Z. (2010). Potential long-term storage of the predatory mite *Phytoseiulus persimilis*. *BioControl, 55*, 639–644.

Şenal, D., Karaca, İ., & Ündağ, H. (2001). Storage possibilities of scale insect predator *Chilocorus bipustulatus* (L.) eggs at different temperatures. *Boll. Zool. Agr. Bachic., Ser. II, 33*(3), 427–433.

Stathas, G. J. (2000a). The effect of temperature on the development of the predator *Rhyzobius lophantae* and its phenology in Greece. *BioControl, 45*, 439–451.

Stathas, G. J. (2000b). *Rhyzobius lophantae* prey consumption and fecundity. *Phytoparasitica, 28*, 203–211.

Stathas, G. J., Eliopoulos, P. A., Kontodimas, D. C., & Siamos, D. T. (2002). Adult morphology and life cycle under constant temperatures of the predator *Rhyzobius lophantae* Blaisdell (Col., Coccinellidae). Anz. Sch dlingskunde/J. *Pest Science, 75*, 105–109.

Tauber, M. J., & Tauber, C. A. (1978). Evolution of phenological strategies in insects: A comparative approach with ecophysiological and genetic considerations. In *Evolution of Insect Migration and Diapause, Proceedings in Life Science* (pp. 53–71).

Tauber, M. J., Tauber, C. A., & Gardescu, S. (1993). Prolonged storage of *Chrysoperla carnea* Steph. (Neuroptera: Chrysopidae). *Environmental Entomology, 22*, 843–848.

Uygun, N. (1981). Taxonomic research on Coccinellidae (Coleoptera) fauna of Turkey. Publications of Ç.Ü. Agricultural Faculty: 157, Scientific Research Thesis: 48.

Uygun, N. (1991). Biological control in Turkey. *Tarım Orman Köyişleri Bak. Derg., 63*, 28–30.

Uygun, N., Ulusoy, M. R., & Satar, S. (2010). Biological control. *Türkish Journal of Biological Control, 1*(1), 1–14.

Yiğit, A., Canhilal, R., & Zaman, K. (1994). *Cold storage of some natural enemies of citrus mealybug, Planococcus citri (Risso) (Homoptera: Pseudococcidae).* Bulletin of III. Turkey Biological Control. Entomological Society Publications, no:7, pp. 137–146.