J. Agr. Sci. Tech. (2015) Vol. 17: 263-273

Demographic Analyses of Resistance of Five Varieties of Date Palm, *Phoenix dactylifera* L. to *Ommatissus lybicus* De Bergevin (Hemiptera: Tropiduchidae)

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ABSTRACT

Demographic analyses of dubas bug, *Ommatissus lybicus* de Bergevin, (Hemiptera: Tropiduchidae), reared on five date palm varieties (Zahdi, Mazafati, Piarom, Khasi, and Shahani) were constructed under laboratory conditions at 27±1ºC, 70±5% RH, and a 14:10 hours (L: D) photoperiod. Data were analyzed based on the age-stage, two-sex life table theory to take the variable developmental rate among individuals and both sexes into consideration. Results showed that different varieties of date palm influenced development time and fecundity of dubas bug. The total pre-adult developmental time was the shortest on Zahdi (85.21 days) and the longest on Khasi (88.39 days). The longevities of both male and female adults on different varieties were significantly different. The mean fecundity per female was significantly higher on Zahdi (78.62 eggs) and Mazafati (68 eggs) than on Shahani (43.53 eggs) and Khasi (46.32 eggs). The highest and lowest net reproductive rates were obtained on Zahdi (40.58 offspring per individual) and Shahani (16.88 offspring per individual), respectively. The intrinsic rates of increase were 0.0401, 0.0368, 0.0355, 0.0312 and 0.0301 per day on Zahdi, Piarom, Mazafati, Khasi and Shahani, respectively. The mean generation times ranged from 92.78 days reared on Zahdi to 95.25 days on Mazafati. According to results obtained in this study, Shahani and Khasi were the most resistant varieties while Zahdi was the most susceptible variety to dubas bug among the varieties tested.

Keywords: Demography, Date palm variety, Dubas bug.

[DOR: 20.1001.1.16807073.2015.17.2.1.4]

INTRODUCTION

The dubas bug, *Ommatissus lybicus* de Bergevin (Hem.: Tropiduchidae), is one of the most important pests of date palm in Iran (Gharib, 1966) and several countries in the Middle East and North Africa (Hussain, 1963; Gharib, 1966; Bitaw and Ben-Saad, 1990). *Ommatissus lybicus* causes crop losses through direct feeding and honeydew contamination of plant parts. Severe infestations reduce vigor and productivity of the palm and cause the fruits to shrink, rendering them unmarketable (Hussain, 1963). This pest has two generations per year and overwinters as egg stage (Hussain, 1963; Klein and Venezian, 1985).

The life table parameters provide crucial information to study population dynamics and to establish management tactics in pest control (Maia *et al.*, 2000). Comparison of life table parameters of an insect pest on different host plant varieties can be used to select resistant varieties (Razmjou *et al.*, 2006; Golizadeh *et al.*, 2009; Razmjou *et al.*, 2009; Obopile and Ositile, 2010; Naseri *et al.*, 2011; Khanamani *et al.*, 2013; Goodarzi *et al.*, 2015). Life table of dubas bug at different temperatures has been studied using traditional female age-specific life table methods by Payandeh *et al.* (2010).

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However, the theories relating to female age-specific life tables (Lewis, 1942; Leslie, 1945; Birch, 1948) ignore the male population and the stage differentiation. An age-stage, two-sex life table theory was developed to resolve these problems by including both sexes and the variable developmental rates among individuals (Chi and Liu, 1985; Chi, 1988).

Although there are several studies related to control measures of dubas bug (Lashkari *et al.*, 2008; Hussain, 1963), no information is currently available on the effect of different host plant varieties on the life table and population growth potential of this pest. The aim of this study was to use an age-stage, two-sex life table theory (Chi and Liu, 1985; Chi, 1988) to determine the impact of five important date palm varieties commercially cultivated in Fars Province, Iran, on the life table and population growth parameters of dubas bug. Knowledge of the life table parameters of dubas bug and resistance-susceptibility characteristics of date palm varieties could be extremely valuable for development of IPM programs against *O. lybicus*.

MATERIALS AND METHODS

This research was conducted in the insectarium of Agricultural Organization of Darab (located in south of Fars Province, Iran) at 27±1°C, 70±5% RH, and a photoperiod of 14:10 (L:D) hour.

Host Plant Varieties and Leafhopper Colony

Five commercial date palm varieties including Zahdi, Piarom, Shahani, Mazafati, and Khasi were tested in this study. The varieties were obtained from Fars Research Center for Agriculture and Natural Resources as offshoots and were then planted in plastic pots (Height: 38 cm, Top diameter: 38 cm, Bottom diameter: 27 cm) filled with suitable field soil. The potted plants were maintained in the insectarium, irrigated weekly and fertilized with HORTI® micro and macro elements fertilizer monthly for one year to be established completely and be suitable for laboratory experiments. For each variety, 7 trees were used. The experiments began in late May 2011 while the adult leafhoppers of the first generation were appearing in nature.

The adults of dubas bug were collected by aspirator from a date palm orchard (variety Khasi) in Darab region, Iran, and were colonized on potted palms. The leafhoppers were reared on related host plant varieties for one generation before being used for the life table experiments. Males and females were differentiated by female's black ovipositor.

Life Table Study

In this experiment, leaflet cages on potted trees were used to measure biological characteristics of dubas bug on each variety. The cage consisted of transparent plastic cylinder (25 mm in diameter and 185 mm in height) which enclosed a single leaflet of the potted trees. Three mesh-covered openings were made on the sides of the cage for ventilation. The openings around the top and bottom of the cage were sealed with a piece of foam to prevent the escape of insects.

In order to obtain the same aged eggs of dubas bug on each variety, three pairs of three-day-old adults were released in the leaflet cages for 24 hours. In total, fifty eggs were used as cohorts for the life table experiments. All the eggs were observed daily and the incubation period was recorded. The emerged nymphs were individually transferred to new leaflet cages using a fine brush. Observations were made daily to record the development times and survivorship of the nymphs. Exuviae from moulting were used to discriminate the nymphal instars. When an individual developed to the adult stage, it was paired with an individual of the opposite sex from the cohort. An aspirator was used to transfer

[DOR: 20.1001.1.16807073.2015.17.2.1.4]

the adult leafhoppers to new cages every day. The daily survival and fecundity were recorded for each individual until death.

Statistical Analysis

The life history raw data were analyzed based on the theory of the age-stage, two-sex life table (Chi and Liu, 1985; Chi, 1988). The computer program TWOSEX-MSChart (Chi, 2005) was used for data analysis. The program is available at http://140.120.197.173/Ecology/prod02.htm (Chung Hsing University) and http://nhsbig.inhs.uiuc.edu/wes/chi.html (Illinois Natural History Survey). The age-stage specific survival rate (S_{xj}, where x is the age and j is the stage), the age-stage specific fecundity (f_{x7}), the age-specific survival rate (l_x), the age-specific fecundity (m_x), and the population parameters (r_m: The intrinsic rate of increase; λ: The finite rate of increase; GRR: The gross reproductive rate; R_0: The net reproductive rate, T: The mean generation time) were calculated accordingly.

Intrinsic rate of increase was estimated using the iterative bisection method from the Euler-Lotka formula, with age indexed from 0 (Goodman, 1982):

$$\sum_{x=0}^{\infty} e^{-r_m(x+1)} l_x m_x = 1 \tag{1}$$

The mean generation time is defined as the period of time that a population needs to increase to R_0-fold of its size at the stable age-stage distribution. The mean generation time is calculated as $T = (\ln R_0)/r$. The means and standard errors of life table parameters were estimated using Jackknife method (Sokal and Rohlf, 1995). Effect of host plant varieties on the population parameters, development times, and fecundities of dubas bug were analyzed using one-way analysis of variance and the means separated by Tukey's honestly significant difference (HSD) test (SPSS, 1999).

RESULTS

Developmental times for each stage of dubas bug on five date palm varieties are shown in Table 1. Mean incubation period on Zahdi was significantly lower than the other four varieties ($F= 8.8$; $df= 4, 189$; $P< 0.05$). Hatching rates on Zahdi, Mazafati, Piarom, Khasi and Shahani were 100, 92.50, 100, 93.62, and 93.88%,

Table 1. Basic statistics (Mean±SE) of the life history of *Ommatissus lybicus* reared on five date palm varieties.[a]

Statistics	Date palm variety: Zahdi	Mazafati	Piarom	Khasi	Shahani
	Developmental time (d)				
Egg	39.1±0.23a	41.35±0.36b	41.25±0.28b	41.7±0.32b	41.13±0.34b
1st instar	4.41±0.09a	4.44±0.08a	4.47±0.087a	4.5±0.082a	4.39±0.07a
2nd instar	7.07±0.09a	7.28±0.1ab	7.52±0.08b	7.35±0.08ab	7.3±0.09ab
3rd instar	8.97±0.11a	9.03±0.12a	9.09±0.126a	9.08±0.101a	9.18±0.12a
4th instar	11.66±0.13a	11.77±0.1a	11.88±0.09a	11.94±0.08a	11.55±0.14a
5th instar	14±0.11b	13.79±0.14ab	13.97±0.10b	13.61±0.11ab	13.42±0.11a
pre-adult	85.21±0.31a	87.94±0.46b	88.12±0.26b	88.39±0.38b	87.21±0.49b
	Adult longevity (d)				
Male	15.25±1.15ab	16.59±0.73b	14.07±1.07ab	12.5±1.14a	11.42±0.89a
Female	17.56±1.21ab	20.47±0.93b	14.39±1.07a	13.37±1.15a	13.63±1.13a
Fecundity (Eggs/Female)	78.62±5c	68±2.74c	63.33±5.45bc	46.32±6.21ab	43.53±4.46a

[a] Means in a row followed by a common letter are not significantly different (P> 0.05; Tukey's HSD).

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respectively.

Total developmental time for all pre-adult stages was the shortest on Zahdi and the longest on Khasi, and the difference between those varieties was significant ($F= 8.78$; $df= 4, 161$; $P< 0.05$). However, differences in the duration of individual nymphal instars were not always significant. The insect passes through five nymphal instars before reaching adult stage.

The longevities of both male and female adults on different varieties were significantly different ($F= 4.90$; $df= 4, 72$; $P< 0.05$ and $F= 7.55$; $df= 4, 84$; $P< 0.05$, respectively) and the longest longevities for both male and female adults occurred on variety Mazafati while the shortest occurred on Shahani and Khasi, respectively (Table 1). The total fecundity varied significantly among varieties ($F= 8.74$; $df= 4, 84$; $P< 0.05$) (Table 1). Significantly higher fecundities were observed in females reared on Zahdi and Mazafati compared to those reared on Shahani and Khasi. Sex ratio (proportion of females in second generation) on different varieties ranged from 50 to 58%.

Figure 1 illustrates the age-stage specific survival rates (S_{xj}) of dubas bug on five date palm varieties. The S_{xj} shows the probability that a newborn individual will survive to age x and stage j. In general, the survival rates of dubas bug on varieties Shahani and Khasi were lower than those found on the other three varieties, and this was due to the higher mortalities of immature stages that occurred on these two varieties. Survival rates from egg to adult emergence on varieties Khasi and Shahani were 0.70 and 0.77, respectively, while on Mazafati, Zahdi and Piarom were 0.85, 0.90 and 0.92, respectively. There were prominent overlaps in survival curves of different life stages. Adult females had more survival rate than males on all varieties and the maximum lifespan (119 days) was observed on variety Mazafati (Figure 1).

Age-stage specific fecundity (f_{xj}) is defined as daily number of eggs produced per female of age x. Because only females could produce egg, the f_{xj} can also be called female age-specific fecundity and shown as f_{x7} (i.e. the adult female is considered as seventh life stage). The age-specific fecundity (m_x) equals the daily number of eggs divided by all individuals (male and female) of age *x*.

Figure 2 shows that the reproduction periods of dubas bug on different varieties were partly similar in pattern as started in the range of 83 to 86th day of leafhopper longevity and peaked 5 to 8 days later. After the peak, mean daily fecundity (f_{x7} or m_x) gradually declined on all varieties tested, as females aged. The maximum f_{x7} (8.19 eggs female^{-1} day^{-1}) was found on variety Zahdi, followed by 6.70, 6.55, 5.19 and 4.88 eggs female^{-1} day^{-1} on Mazafati, Piarom, Khasi and Shahani, respectively.

The age-specific survival rate (l_x) is also presented in Figure 2. The l_x curve ignores male adults and the stage differentiation among individuals, therefore, it is a simplified form of S_{xj}.

The Population parameters of dubas bug including intrinsic rate of increase (r_m), finite rate of increase (λ), gross reproductive rate (GRR) , net reproductive rate (R_0) and mean generation time (T) on five date palm varieties are presented in Table 2. The maximum value of r_m was recorded for leafhoppers reared on variety Zahdi, and this value was significantly different from those obtained on both Khasi and Shahani ($F= 3.33$; $df= 4, 198$; $P= 0.012$). In addition, there was no significant difference in r_m values between Piarom and Mazafati, or among these two and other varieties tested (Table 2). A similar pattern was found for parameters λ and R_0 (Table 2). However, no differences were observed in *GRR* or *T* for dubas bug reared on different varieties ($F= 1.47$; $df= 4, 198$; $P= 0.212$ and $F= 1.79$; $df= 4, 198$; $P= 0.133$, respectively) (Table 2).

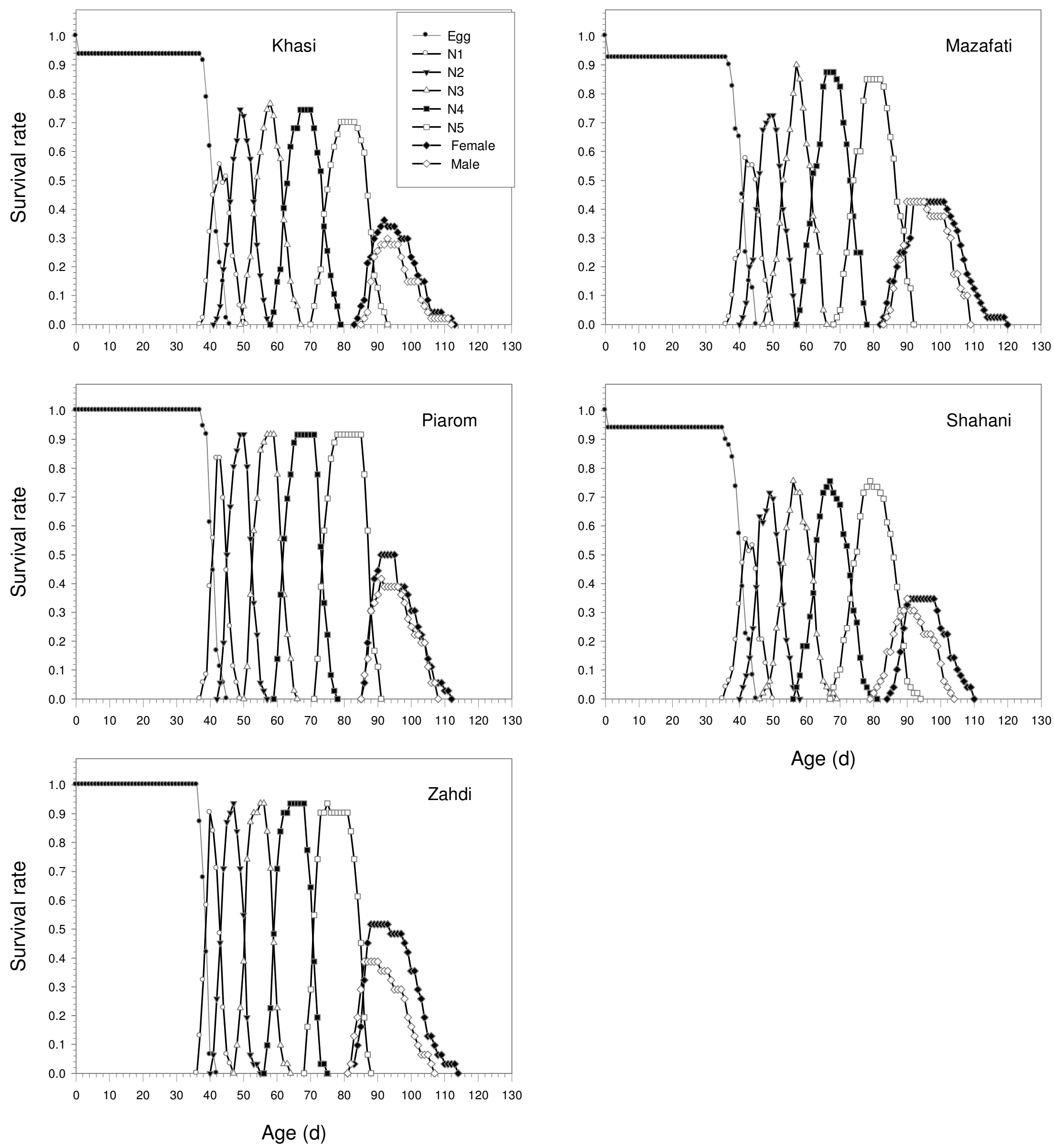

Figure 1. Age-stage specific survival rate (S_{xj}) of *O. lybicus* on five date palm varieties.

Table 2. Mean±SE of intrinsic rate of increase (r_m) (day^{-1}), finite rate of increase (λ) (day^{-1}), gross reproductive rate (GRR) (offspring per individual), net reproductive rate (R_0) (offspring per individual), and mean generation time (T) (days) of *Ommatissus lybicus* on five date palm varieties.[a]

Population parameters	Date palm variety					
	Zahdi	Mazafati	Piarom	Khasi	Shahani	P
r_m	0.0401±0.002b	0.0355±0.0021ab	0.0368±0.002ab	0.0312±0.0024a	0.0301±0.0022a	0.012
λ	1.0409±0.0021b	1.0361±0.0021ab	1.0375±0.0021ab	1.0317±0.0024a	1.0306±0.0023a	0.012
GRR	55.42±9.42a	50.89±10.61a	40.51±7.37a	37.37±8.11a	30.84±5.34a	0.212
R_0	40.58±7.61b	28.9±5.5ab	31.67±5.99ab	18.72±4.16a	16.88±3.5a	0.010
T	92.78±0.5a	95.25±0.81a	94.44±0.49a	94.82±0.7a	94.5±0.53a	0.133

[a] Means in the same row followed by the same letter are not significantly different (P> 0.05; Tukey's HSD).

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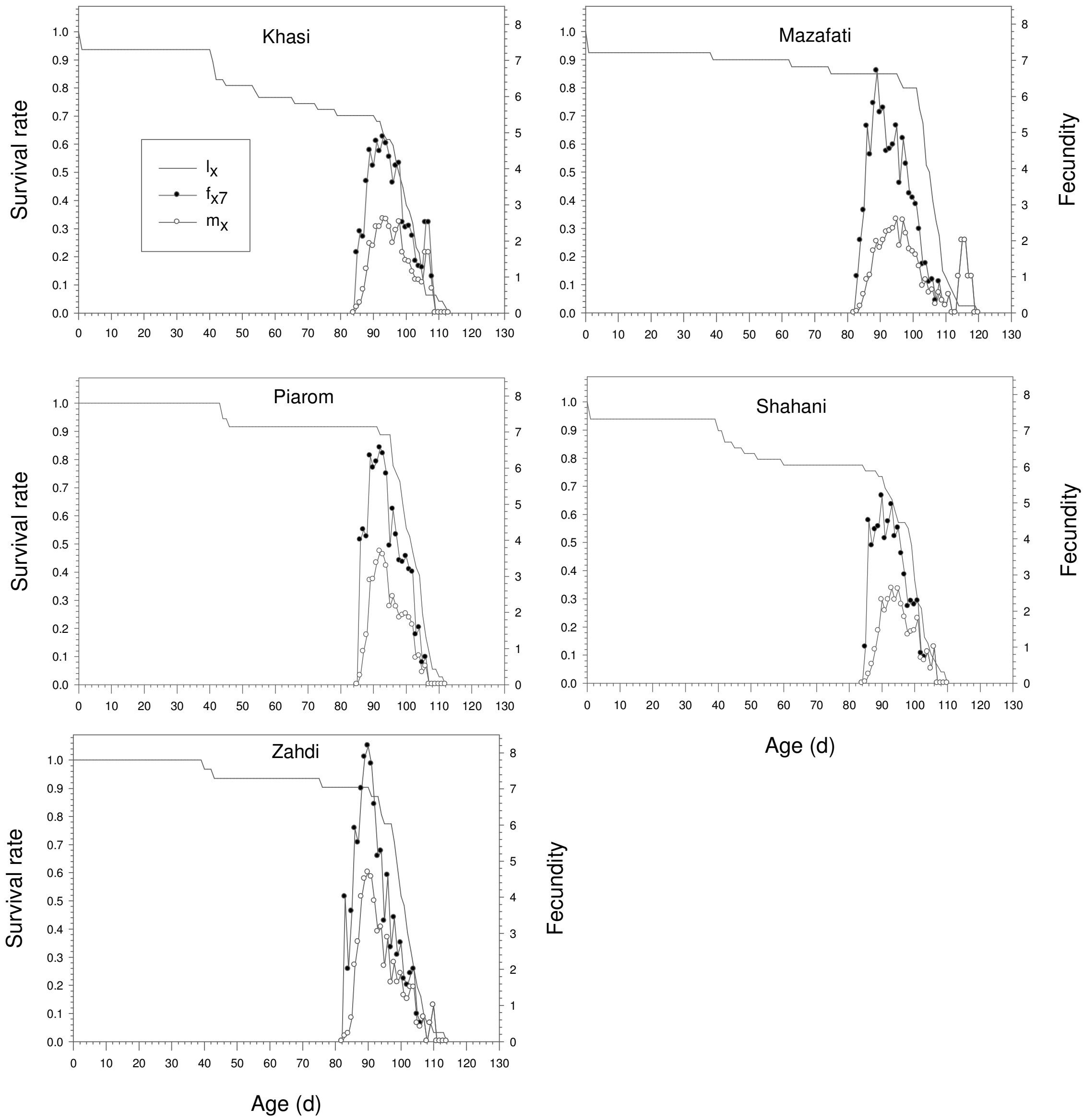

Figure 2. Age-specific survival rate (l_x), female age-specific fecundity (f_{x7}) and age-specific fecundity of total population (m_x) of *O. lybicus* on five date palm varieties.

DISCUSSION

Results of this study showed that different varieties of date palm can significantly affect development time, survival and fecundity of dubas bug. Egg incubation period of leafhoppers reared on Zahdi was significantly lower than the other varieties. This might be caused by the different food sources taken up by the parents before oviposition and subsequent effect on embryonic development. Similar inference has been reported for *Plutella xylostella* (L.) reared on five cultivated brassicaceous host plants (Golizadeh *et al.*, 2009) and for *Copitarsia decolora* Hampson reared on asparagus and artificial diet (Gould *et al.*, 2005).

Despite the importance of plant variety effects on population growth of pests, there was no study available regarding the effect of date palm varieties on dubas bug life table. However, there were two studies that documented the effect of different constant temperatures on age-specific life table (Payandeh *et al.*, 2010) and on pre-adult development time of dubas bug (Mokhtar and Nabhani, 2010). Our results regarding the egg incubation period, which ranged from 39.1 to 41.7 days, are definitely longer than those reported by Payandeh *et al.* (2010) at 25ºC (32.41 days) or 30ºC (30.89 days). On the other hand, this period is somewhat close to the corresponding value at 27.5ºC (39.2 days), and very shorter at 25 and 30ºC (47.6 and 50.8 days, respectively) in Mokhtar and Nabhani (2010).

The total developmental time for all pre-adult stages ranged from 85.21days on Zahdi to 88.39 days on Khasi. This range is longer than those reported by Payandeh *et al.* (2010) at 25ºC (82.48 days) and 30ºC (75.79 days), but is close to those reported by Mokhtar and Nabhani (2010) at 27.5ºC (83.9 days) and 30ºC (88.4 days). However, the authors have not cited the name of date palm variety used in their study.

The longevities of male and female adults were also variety-dependent and the longest longevities for both male and female adults occurred on variety Mazafati (16.59 and 20.47 days, respectively), which were close to those reported by Payandeh *et al.* (2010) at 30ºC (15.61 and 19.79 days, respectively).

Differences among varieties in developmental times and adult longevities could be attributed to plant differences in nutritional content and/or morphological characteristics. However, the variations between results of different studies could be result of differences in experimental conditions, especially temperature, and host plant quality. In addition, it seems reasonable to speculate that such pronounced intraspecific variability between dubas bug population in the study by Payandeh *et al.* (2010) and the current study is associated with differences among geographic populations of *O. lybicus*. Similar inference has been made by Nielsen *et al.* (2008) for *Halyomorpha halys* (Stal), Auad *et al.* (2009) for *Rhopalosiphum padi* (L.) and Golizadeh *et al.* (2009) for *Plutella xylostella* (L.). When an insect species has a wide geographic distribution, it would be expected that intraspecific variation occurs among its populations (Diehl and Bush, 1984). Geographic variation in biological traits of some insects, especially parasitoids, has already been reported by several authors (van den Bosch *et al.*, 1979; Claridge *et al.*, 1982; Awan *et al.*, 1990; Liu *et al.*, 2001).

The type of analytical methods and life table theories used to describe demography can also affect the results of life table studies (Chi, 1988; Jha *et al.*, 2012). In the present study, we used age-stage, two-sex life table which has distinct advantages over traditional age-specific life table. For example, the age-stage, two-sex life table takes variable development rate among individuals into account while in age-specific life table the differences in pre-adult development are ignored, and it is assumed that all adults emerge on the same day. This assumption will not only inaccurately diminish the real variability among individuals, but also consequently results in miscalculations of the survival and fecundity curves (Chi, 1988; Chi and Yang, 2003; Yu *et al.*, 2005; Chi and Su, 2006; Kavousi *et al.*, 2009; Huang and Chi, 2012). Chi and Yang (2003), Chi and Su (2006), and Kavousi *et al.* (2009) gave detailed explanations and mathematical proofs to demonstrate the errors in female age-specific life tables.

The intrinsic rate of increase (r_m) was significantly affected by variety of date palm. The highest r_m value was found on Zahdi variety, followed by Piarom, Mazafati, Khasi, and Shahani. These results indicated that Zahdi was the most susceptible to the dubas bug among the varieties tested. In contrast, Shahani variety had adverse effect on population growth, indicating some level of resistance to dubas bug. Intrinsic rate of increase is a reflective of many biological traits of an insect pest including survival, fecundity, development time and sex ratio (Lewontin, 1965; Southwood and Henderson, 2000; Ozgokce and Atlihan, 2005; Fathi, 2009; Golizadeh *et al.*, 2009; Jha *et al.*, 2012). For example, the highest value of *r* on Zahdi was more due to the highest fecundity (78.6 eggs) and the lowest pre-adult development time (85.21 days) among the varieties tested.

The lowest fecundity and survival of dubas bug on Shahani and Khasi, as reflected in *r*, may possibly be attributed to the presence of antibiosis in these varieties. However, in addition to the life table assay, there is a need for host preference assays to characterize any antixenotic effects. Plant resistance mechanisms include chemical defenses (antibiosis) that influence biological processes of insects, like survival, generation time, fecundity, and longevity (van Emden, 1997) and morphological defenses (antixenosis) that deter insects from oviposition, feeding and colonization (Saxena, 1990; van den Berg and van der Westhuizen, 1997; Reddy *et al.*, 2002).

Similar to the results for the intrinsic rate of increase, the values of net reproductive rate (R_0) and finite rate of increase (λ) indicated that the variety Zahdi was the most favorable host for dubas bug compared to the other tested varieties. However, the gross reproductive rate (GRR) and mean generation time (T) did not differ significantly among the tested varieties. Because the *GRR* ignores the age-specific survival rate, its ecological significance and the result of statistical comparison should be interpreted with caution (Yu *et al.*, 2005; Chi and Su, 2006).

As proven by Chi (1988), for a two-sex population, the relationship between the net reproductive rate (R_0) and the mean female fecundity (F) is:

$$R_0 = \frac{N_f}{N} \cdot F \qquad (2)$$

Where, N is the total number of eggs used at the beginning of the life table study, and N_f is the number of female adults emerging from these N eggs. All of our results for dubas bug on different varieties were consistent with the relationship of Equation (2). If a life table is constructed based on age-specific female life table, the errors in survival rate (l_x) and the fecundity (m_x) will finally result in errors in R_0, and the relationship between R_0 and F will be inconsistent with the Equation (2) (Chi, 1988; Chi and Su., 2006)

In the current study, the calculated values of r, R_0, λ and *GRR* on all five varieties of date palm were lower than those reported by Payandeh *et al.* (2010) at both 25 and 30ºC. As discussed above, the difference between our results and those reported by Payandeh *et al.* (2010) may be due to differences in experimental conditions, plant varieties, geographic populations, analytical methods, and life table theories.

Knowledge of the extent of susceptibility or resistance of plant varieties to insect pests is a fundamental component of integrated pest management (IPM) programs. According to results obtained in this study, Shahani and Khasi were the most resistant varieties while Zahdi was the most susceptible variety for dubas bug among the varieties tested. Since Shahani and Khasi are somewhat locally specific varieties, it seems they can be recommended mainly in Fars and Hormozgan Provinces. However, the resistant varieties found in this study can constitute a useful genetic resource for date palm breeding programs aimed at developing resistant varieties against dubas bug. For further study, examining the effect of chemical compound or morphological structure of these varieties on population growth of dubas bug and other date palm pests will be worthy of pursuit. In addition, comparative studies that include different geographic sources of dubas bug population would be useful to reveal intraspecific variations which, if existing, could be considered for more precise management programs against the pest.

[DOR: 20.1001.1.16807073.2015.17.2.1.4]

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[DOR: 20.1001.1.16807073.2015.17.2.1.4]

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بررسی مقاومت پنج رقم نخل خرما *Phoenix dactylifera* L. به زنجرک خرما Ommatissus lybicus de Bergevin (Hemiptera: Tropiduchidae)با استفاده از تجزیه وتحلیل دموگرافیک

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چکیده

تأثیر پنج رقم نخل خرما (زاهدی، مضافتی، پیارم، خاصی و شاهانی) روی ویژگی های دموگرافیک زنجرک خرما، Ommatissus lybicus de Bergevin، در دمای ۱ ± ۲۷ درجه ی سلسیوس، رطوبت نسبی ۵ ± ۷۰ درصد و دوره‌ی نوری ۱۴ ساعت روشنایی و ۱۰ ساعت تاریکی تعیین شد. برای در نظر گرفتن هر دو جنس نر و ماده و همچنین نرخ رشد و نمو متغیر میان افراد در محاسبه ویژگی های دموگرافیک، داده‌ها بر اساس تئوری جدول زندگی سنی-مرحله رشدی دوجنسی تجزیه و تحلیل شدند. نتایج نشان داد ارقام مختلف خرما دارای اثر متفاوتی بر طول دوره رشد و نمو و باروری زنجرک خرما دارند. کوتاه‌ترین و طولانی‌ترین دوره قبل از بلوغ به ترتیب ۸۵/۲۱ و ۸۸/۳۹ روز روی ارقام زاهدی و خاصی بود. ارقام مختلف دارای اثر معنی داری بر طول عمر زنجرک‌های نر و ماده بالغ داشتند. زنجرک هایی که روی ارقام زاهدی و مضافتی پرورش یافتند به طور معنی داری میانگین باروری (تخم/ماده) بیشتری نسبت به ارقام شاهانی و خاصی داشتند. بیشترین و کمترین مقدار نرخ خالص تولیدمثل به ترتیب روی ارقام زاهدی (۴۰/۵۸ نتاج/فرد) و شاهانی (۱۶/۸۸ نتاج/فرد) بود. نرخ ذاتی افزایش جمعیت روی ارقام زاهدی، پیارم، مضافتی، خاصی و شاهانی به ترتیب ۰/۰۴۰۱، ۰/۰۳۶۸، ۰/۰۳۵۵، ۰/۰۳۱۲ و ۰/۰۳۰۱ در روز بود. میانگین طول نسل زنجرک خرما بین ۹۲/۷۸ تا ۹۵/۲۵ روز به ترتیب روی ارقام زاهدی و مضافتی متغیر بود. بر اساس نتایج حاصل، ارقام شاهانی و خاصی به عنوان گیاهان میزبان مقاوم‌تر و رقم زاهدی به عنوان میزبان حساس‌تر برای زنجرک خرما شناخته شدند.

[DOR: 20.1001.1.16807073.2015.17.2.1.4]