J. Agr. Sci. Tech. (2012) Vol. 14: 257-266

A Study on Some Biological Characteristics of Olive Leaf Moth, *Palpita unionalis* Hübner (Lep: Pyralidae) in Iran

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ABSTRACT

Nearly just a decade ago, olive leaf moth, *Palpita unionalis*, was identified as an exotic pest from an olive grove in Roodbar, Iran. In order to plan an IPM programme regarding olive, a series of laboratory and field studies were carried out during 2001-2003 in Qazvin Province. To study the emergence time, number of generations, feeding behaviour and developmental periods at field, five trees (Zard cultivars) were selected within two groves, twenty infested branches of which (4 per tree) were taken to be covered with fine mesh net. They were monitored weekly. In the laboratory, a number of the pest pupae were taken to predefined conditions and reared at 25±0.5°C, 65±5% RH and 12:12 L:D. Then, the developmental period at various life stages, adults' life span, females' fecundity as well as sex ratios were determined. The field results showed that it took 34.2±2.1 days from neonate larval stage to adult emergence. Moreover, it was revealed that the pest had 4 to 5 overlapping generations per year. Based on the laboratory experimental results, embryonic developmental time, whole larval stages, pupal duration and female and male longevity took 5.8±1.08, 21.6±0.33, 8.33±1.0 and 12.6±1.3, and 13.5±1.1 days, respectively. Besides, females laid 182.33±18.1 eggs, on the average.

Keywords: Biology, Olive leaf moth, *Palpita unionalis*, Qazvin (Tarome–Sofla).

INTRODUCTION

Olive leaf moth is one amongst the important olive pests prevailent in Egypt, Greece, and Italy (El-Kifl *et al*., 1974; Longo, 1992; Pertich, 1988; Vassilaina-Alexopoulou and Santorini, 1973). Various aspects of the pest have been investigated in different countries. The pest has also been reported on a range of other plant hosts including *Ligustrum vulgare*, *Arbutus unedo*, *Fraxinus* spp., and *Pillyrea media* (Mazomenos *et al*., 2002).

Shehata *et al*. (2003) studied the pest biology in Egypt. Azimizade *et al.* (2003) investigated *Ligustrum vulgare* as a desirable host for breeding of the pest in laboratory in comparison with Olive cultivars.

An extensive study on the pest biology (Grossley, 2000) revealed that Olive Leaf Moth (OLM) sustained five instars larvae. It was also revealed that the egg incubation period lasts 3.5 days at 15-25°C temperatures (Gargani, 1999). Pinto (1995) determined olive leaf moth as a major pest causing extensive damage to young olive trees in a sapling nursery, while studying morphology and biology of Pyralids. Pinto and Salerno (1994) did some research on the pest's growth and developmental stages, longevity, parthenogenesis, search for host and ovipositing during which they also recorded *Apantles syleptae* as one of its effective parasitoids. Similarly, Khaghaninia (2002) recorded a tachinid parasitoid, *Carcelia* sp, on the pupa of OLM in the Tarome Olia region.

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Studies conducted by Athanassiou *et al.* (2004) on the male trapping techniques at field, revealed that the percentage rate of males trapped increased drastically in fall as compared with the capture rates during the summer.

Kirti and Rose (1992) prepared an identification key for different species of *P. unionalis* based on the internal and external differences of male genitalia in India. Longo (1992) suggested an integrated management strategy with regard to the pest. Loi (1990) evaluated the effect of different temperatures, in a range of 10-35°C on OLM embryonic growth and development.

In Iran, it took only a few years for the Olive Leaf Moth (OLM) *Palpita unionalis*, to spread as a serious pest throughout the country since its first being reported by Saeb (1999) from Roodbar, Guilan, in August, 1999. It is now a significant pest of nurseries and young trees in olive groves in the provinces of Mazandaran, Zanjan and Qazvin (Pazuki 2000) as well as of Tehran (Ghavami, 2000).

Field observations of the authors indicated that normally the first adult moths emerge from their over-wintering larvae late in February with the first generation being completed by the end of March and in early April. The second generation sets in around mid May, depending on the prevailent climatic conditions. However, the pest population reaches its peak during its third and forth generations, i.e., in mid June and first half month of July, while decreasing gradually (Figure 1).

The adult moth is white in color with a wing span of about 29 mm. Front wings are wider than the hind ones. Anterior margin of wings are brown, bearing two black spots in the middle. The wings bear frenolum, and in resting position stand on the body in gable roof form. The males and females do not differ in length and width. Females possess a mating pore on the eighth segment of the abdomen and an ovipositing pore on the ninth one. Females' abdomen is light green covered with white scales, but in males there is a pore on that terminal part of the body that bears sets of hair. The adults are active early in the morning or during sunset while exhibiting a low level of activity, possibly with short flights during the warmer hours of the day. Adults usually mate one to two days after emergence and oviposit within three to seven days. Eggs are flat oval, light greenish yellow, 1.02 mm long and 0.49 mm wide, exhibiting a mesh appearance and usually seen sporadically on the underside of leaves. Larva is eruciform having three thoracic and five abdominal legs on the 3, 4, 5, 6, and 10^{th} abdominal segments. Crochets are seen in closed and complete spherical form at the

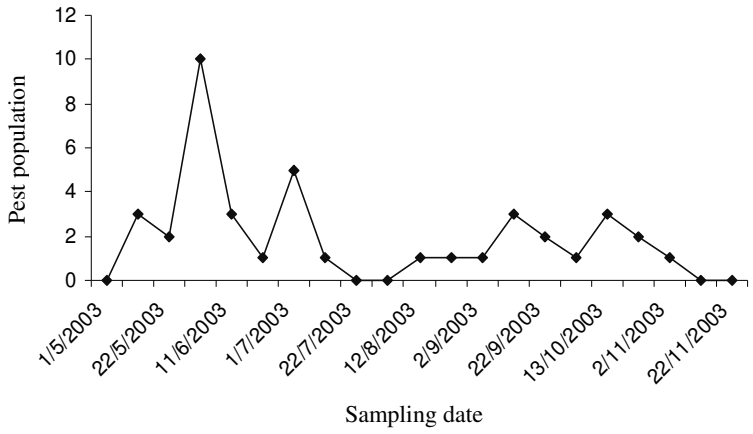

Figure 1. Population fluctuations of olive leaf moth, *Palpita unionalis*, in Tarome Sofla, Qazvin.

end of prolegs. Larvae are pale yellow in 1st and 2nd instars becoming green in later instars. Sometimes a pair of black spots are seen on the body segments close to the pleural part at 3rd and 4th instar stages.

Although, there are nowadays ever increasing reports of OLM, yet, there is no precise remedy measure reported towards its control. On the other hand, the National Plan of Olive Development Plantations for the Purpose of Oil Self-sufficiency in the country justifies a rational plan for its pest control. Therefore, to plan an IPM programme for OLM control, a research was taken up to elucidate the biological characteristics of *P. unionalis* in one of the pivotal olive producing provinces of the country plantations.

MATERIALS AND METHODS

The study was carried out at field and as well under laboratory conditions.

Field Studies

The field experiments were conducted in two groves, Tarome-Sofla, Qazvin. Five plots each containing 5 trees of the same variety were selected in each grove. To determine the time of adult moths emerging from their overwintering larvae in the field, 20 infected shoots (4 per tree) were enclosed in fine-mesh muslin nets in each one of the groves, and weekly monitored. The time from neonate larvae to the next adult moths was also assessed in the same way. Moreover, weekly sampling and counting of the moths at field conditions were done through 500 beatings to randomly selected branches inside a net to specify the pest's status as well as generation throughout the season. As for the second year, the pest was studied *in situ* during the winter season. The OLM wood-boring feeding behaviour was investigated by enclosing the trunk of 20 selected trees in each grove and monitoring them weekly. The methods of field observation and sampling were mainly based on Fairclough (1981) along with some modifications.

Laboratory Studies

To determine the ovipositing time and site(s), fecundity and adults' life span, a number of pupae were collected from field and incubated at 25±0.5°C, 65±5% RH and 12:12 L:D. Ten pots (16 cm in diameter), each containing 1-2 year old saplings, enclosed in nets of 1.5 m heights were prepared (Kumral *et al.,* 2007). Two pupae (1 male and 1 female) were released onto each plant and monitored daily. To determine the duration of larval and pupal stages, ten pots each bearing ten 1st instar larvae were monitored daily. Larval and pupal periods as well as the sex ratios of adults were examined through observations. The larval stages were differentiated as based on the size of larval head capsule (Vassilaina-Alexopoulou and Santorini, 1973). The effect of different densities (ratios) of male and female (1:1, 1.5:1 and 1:1.5 m:f) on the fecundity of females was investigated by confining the related ratios of peers and measuring their daily fecundity till they all died.

Statistical Method

The main experiments were conducted using Complete Randomized Design (CRD). The data in all tests were collected and arranged in Excel worksheets for later analysis. Then ANOVA was carried out through Minitab software. When necessary, data was normalized through proper conversion. Means were separated through Duncan's Multiple Range Test.

RESULTS

Field studies revealed that the pest had 4-5 generations in Qazvin Province environmental conditions (Tarome-Sofla orchards). Approximate time durationof

Table 1. The first larval stage observation per generation of *Palpita unionalis* under field conditions, Tarome Sofla, Qazvin.

Generation	Date of first larva observation
Over-wintering	28/2/2000
First	8/4/2000
Second	15/5/2000
Third	15/6/2000
Fourth	14/7/2000

each generation has been presented in Table 1. The first instar larvae feed on terminal buds while in later instar stages, they move to the lower surfaces of the leaves. Larvae emit silky threads which stick the terminal leaves of shoots to each other and feed in between them. This causes drying up of the terminal buds which stimulates growing of further lateral buds which in turn provides new shoots to be attacked by the next generations. Eventually, branches are unable to grow outside of the tree crown; it turns into a ball form, and the pruning form disrupts completely. In addition, shoots cannot play their role as a flower producer next year. The pest bears five larval instars and over-winters as young larvae between the foliage of the olive trees. Mature larvae spin silky threads and use them to lower themselves to the ground. These larvae make silky cocoons under clods, rubble stones and pupate inside soil, although a few pupae develop between feeding leaves and split in tree trunks. They are brownish green at first, gradually changing into dark brown.

The average duration of larval and pupal stages were determined to be 34.2 ±1.01 days under natural conditions (Table 2). Observations on the confined larvae in the field revealed that the pest did not have a habit of wood-eating in its diet, feeding only on young leaves, even avoiding older leaves that have become coarser than their preferred diet.

The laboratory experiments demonstrated that mating started one day after adult emergence with oviposition occurring two days past mating. The highest oviposition occurred during the first three days, gradually decreasing thereafter (Figure 2). Statistical analysis revealed a significant difference between fecundities on different days ($P< 0.001$) (Table 3). Moreover, females produced 185±19.1, 161±14.2 and 201±21 eggs during the first, second and third years, respectively (Table 7).

The results of m:f ratios on the fecundity experiment proved that the maximum first day oviposition was observed at 1:1 male to female densities as compared with those in other treatments (Figure 3). Similarly, the first day fecundity of 1.5m:1f treatment was higher as compared to that of 1m:1.5f. However, the latter first day females' fecundity was even lower as compared to its 2nd and 3rd days', thereby, for the 1m:1.5f treatment, the highest daily fecundity being observed on the 2nd day. Although there were no significant differences observed among treatments, considering total eggs laid ($\alpha= 0.05$), however, the first day fecundity was statistically different (Figure 3, Table 4).

The findings on the embryonic stage revealed a 5-6 day period as its developmental time (Table 7). Similarly,

Table 2. Larval-adult time duration (Days) of *Palpita unionalis* under field conditions, Tarome Sofla, Qazvin.

Replication	Date of putting larva in cage	Date of first adult emergence	Longevity (Days)
1	17/8/2001	18/9/2001	32
2	17/8/2001	18/9/2001	32
3	17/8/2001	22/9/2001	36
4	17/8/2001	23/9/2001	37
5	17/8/2001	20/9/2001	34
Mean			34.2±1.01

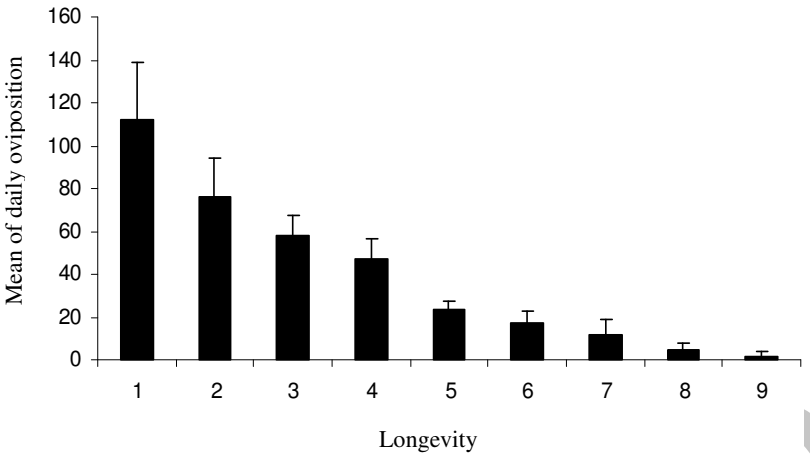

Figure 2. Mean (±SE) daily oviposition per *Palpita unionalis* female at 25±0.5°C, 65±5% RH and 12:12 L:D.

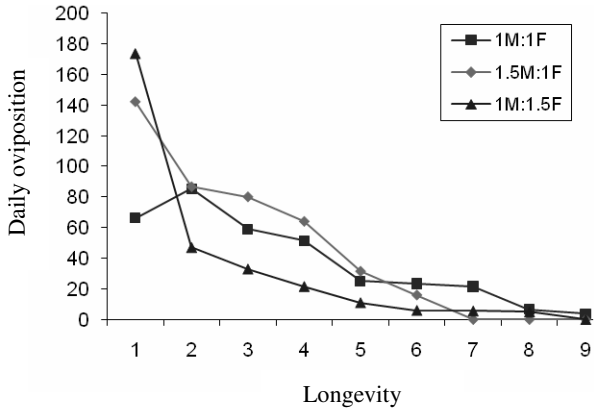

Figure 3. The effect of sexual rate on fecundity of *Palpita unionalis* at 25±0.5°C, 65±5% RH and 12:12 L: D.

Table 2. Larval-adult time duration (Days) of *Palpita unionalis* under field conditions, Tarome Sofla, Qazvin.

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4	17/8/2001	23/9/2001	37
5	17/8/2001	20/9/2001	34
Mean			34.2±1.01

Table 3. Statistical analysis of daily ovipositional fluctuations of *Palpita unionalis*.

S.O.V	df	SS	MS	F	P
Total	35	61576	-		
Treatment	8	43753	5469	9.52	0.01
Error	27	15503	574		

results of larval period experiments indicated five instar larvae for the pest. Table 5 shows the larval stage duration and as well the body size for each instar. Means of larval and pupal stage durations over three years are shown in Table 7.

The larvae fed greedily in their early stages (up to 6-7 days) and then they decreased it till their becoming pupae. Total larval period took about 18-24 days (Tables 5 and 7). One to two days after reaching the last instar, they entered their prepupation phase and finally pupated.

In laboratory conditions, Olive Leaf Moth completed its life cycle within ~32 days. The overall results of 2 year experiments, as presented in Table 6, verified the sex ratio to be surprisingly male based (60%). Moreover, the mean life spans of males and females were 12.1±1.8 and 13.1±0.8 and 13.8±0.7 and 13.2±1.5 for the 1st and 2nd year, respectively (Table7).

Finally, the pest was observed to complete 7 generations under laboratory conditions.

DISCUSSION

Accordng to the results obtained during the present study the pest completed 4-5 generations per year at field conditions. This

Table 4. ANOVA results of sexual rate effect on fecundity of *Palpita unionalis*.

S.O.V	df	SS	MS	F	P
Total	26	5892	-		
Treatment	2	687	343	3.06	Ns
Error	24	25205	1050		

Table 5. Developmental period of *Palpita unionalis* larval instars (days) and their mean (±SE) body size at 25±0.5°C, 65±5% RH and 12:12 L:D.

Larval instar	Larval period (Day)	Larva body size (mm)
First	2-3	3±0.3
Second	5-6	4±1.1
Third	3-4	9±1.0
Fourth	3-4	14±1.2
Fifth	4-5	19±1.1

Table 6. Sexual ratio of *Palpita unionalis* progeny measured in two years reared at 25±0.5°C, 65±5% RH and 12:12 L:D.

Gender	2002	2003	Total
Male	58	71	129
Female	35	50	85
Sexual rate (M:F)	0.62 : 0.38	0.59 : 0.41	0.6 : 0.4

Table 7. Mean (±SE) of embryonic, larval and pupal developmental period, female and male longevity (days), as well as fecundity of *Palpita unionalis* at 25±0.5°C, 65±5% RH and 12:12 L:D.

	Mean±SE			
Stage	2001	2002	2003	Mean
Embryonic period	6 ±0.85	5.4 ±1.2	6 ±1.2	5.8±1.08
Larval period duration	18 ±0.285	22.8 ±0.4	24 ±0.3	21.6±0.33
Pupal period duration	6 ±1.03	10 ±0.77	9 ±1.2	8.33±1.0
Female longevity	-	12.1±1.8	13.1 ±0.8	12.6±1.3
Male longevity	-	13.8 ±0.7	13.2 ±1.5	13.5±1.1
Fecundity	185±19.1	161 ±14.2	201 ±21	182.33±18.1

finding is partially at par with the results concluded by El-Kifl *et al.* (1974), Badawi *et al*. (1976) as well as Fodal and Mule (1990) who reported 10, 9 and 5 generations annually for the pest in some Mediterranean countries. However, Grossley (2000) believes that, the pest has 2-3 generations in cold to mild regions while more than 5-6 in mid-tropical and tropical regions. Therefore, considering the climatic conditions of local distribution of the pest in Iran, the result is very similar to that obtained in this research work (5 generations per year). On the other hand, investigations of Khaghaninia and Farshbaf Pourabad (2009) on the biology of the pest at constantly stable conditions (27 ºC, 65% RH and 16:8 L:D) resulted in 9 generations per year.

The results indicated that the pest could overwinter as 2nd and 3rd instar larval stages which is congruent with the works of Santorini and Alexopoulou (1977), Katsoyannos (1992) as well with that of Triggiani (1971).

Throughout this study, the mean total developmental period for *P. unionalis* reared on Zard olive cultivar, at 25±0.5°C, 65±5% RH and 12:12 L:D was found to be ~32 days. However, there are reports of 27.52-30.00 days of developmental period for *P. unionalis* reared on olive, ash and jasmine under long day lighting, 25ºC and 65% of RH (Kumral *et al.,* 2007), 26 days for the species on olive at 26ºC (Vassilaina-Alexopoulou and Santorini, 1973) and 27 days for the pest on olive at 25 ºC (Shehata *et al.,* 2003). Similarly, El-Khawas (2000) stated that the duration of pre-imaginal development on young olive shoots ranged from 21 to 30 days at 27 ºC and 65% RH. Moreover, Badawi *et al.* (1976) and Arambourg (1986) reported that the duration of post-embryonic development varied from 29.2 to 32.0 days on different host plants including olive, jasmine and ash tree. The differences depicted in these research works including the present one could be due to olive varieties, their ages, environmental conditions under which the experiments performed, as well as the pest biotype.

Adult longevity of *P. unionalis* determined 12.6 and 13.5 days for female and male individuals, respectively. However, these figures seem different from results obtained by Shehata *et al.* (2003) who reported 11.4 and 11.9 days for female and males on olive under similar laboratory conditions.

There have been different reports on the fecundity of the female OLM. The present 3 years of investigation demonstrated that females were capable of laying about 182±18.1 eggs during their whole life. On the contrary, Loi (1990) and Grossley (2000) as well as El-Kifl *et al*. (1974) documented 86-515, 141-882 and 414 eggs per female, respectively. The observed differences in fecundity could be due to feed sources (plant varieties), the pest biotype differences etc. Besides, Badawi *et al*. (1976) studies showed that 60% of eggs were laid in sporadic form. On the other hand El-Kifl *et al*. (1974) reported that some eggs were laid in batches of 2 to 6 eggs. In this study, the eggs were laid on the underside of leaves mainly in sporadic form but sometimes in batches of 3-5 eggs which is at par with the findings of El-Kifl *et al*. (1974).

Mean duration of the embryonic developmental stage was found as 5.8±1 days, while El-Kifl *et al*. (1974) have reported that under suitable temperature conditions, it could be completed within 3-9 days, which on the average is similar to the present study's related figures.

Larval stage duration was determined to be 21.6±0.3 days under laboratory conditions. Similarly, Vassilaina-Alexopoulou and Santorini (1973) estimated it as 21-26 days at 25°C and 65% relative humidity in Sicily. But Fodal and Mule (1990) reported it as 24-30 days, the observed difference for which could be related to pre-pupation period.

El Kifl *et al*. (1974) determined a pre-pupation period of 1-2.5 days and pupation period of 9 days in summer and 17-18 days during the winter. Our experiments confirm their report, but Badawi *et al*. (1976) came up with a pupation period of 17 days at 20°C.

The present experiments' findings on the progeny sex ratio demonstrated a ratio of 0.6:0.4 male:female. In contrast, Fodal and Mule (1990) studies revealed a sex ratio of 1:1.6 (male:female). Similar to the current findings, Vassilaina-Alexopoulou and Santorini (1973) reported that the proportion of males was higher as compared with that of the females.

ACKNOWLEDGEMENTS

The authors are sincerely thankful to Olive Studies Office, Ministry of Jihad-e-Agriculture, Islamic Republic of Iran (MJA, IR) as well to the Iranian Research Institute of Plant Protection, MJA, IR, for the financial and technical support of the project. The authors would also like to thank Mr. David Butcher for his editing of the manuscript.

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مطالعه‌ی برخی خصوصیات زیستی پروانه‌ی برگخوار زیتون *Palpita unionalis* Hübner (Lep: Pyralidae) در ایران

ح. نوری و ج. شیرازی

چکیده

تقریبا حدود یک دهه‌ی قبل پروانه‌ی برگخوار زیتون، *Palpita unionalis*، به عنوان یک آفت وارداتی از باغات زیتون رودبار در ایران گزارش شد. به منظور طراحی یک برنامه‌ی مدیریت تلفیقی در زیتون، طی سال‌های ۱۳۸۰ تا ۱۳۸۳ مطالعاتی در سطح باغ و آزمایشگاه در استان قزوین صورت گرفت. جهت بررسی زمان ظهور، تعداد نسل، رژیم غذایی و دوره رشدی آفت، درهر یک از دو باغ دارای رقم زیتون زرد پنج درخت آلوده انتخاب و ۲۰ شاخه (۴ شاخه در هر درخت) در هر باغ انتخاب و با توری مناسب پوشانیده شد و به طور هفتگی مورد بازدید قرار گرفتند. بعلاوه، بوسیله‌ی پرورش آزمایشگاهی، دوران رشدی مراحل زیستی مختلف آفت، طول عمر، باروری ماده‌ها و نسبت جنسی در شرایط ۰/۵±۲۵ درجه سلسیوس، ۵±۶۵ درصد رطوبت نسبی و دوره‌ی روشنایی ۱۲ و تاریکی ۱۲ ساعت تعیین شد. نتایج نشان داد که میانگین طول دوره رشدی لاروهای نئونات تا حشرات بالغ ۲/۱±۳۴/۲ روز در شرایط باغ بود. بعلاوه در همین شرایط، تعداد نسل آفت بین ۴ تا ۵ نسل مختلط در سال تعیین شد.

همچنین بر اساس مطالعات آزمایشگاهی، طول دوره جنینی تخم، لاروی، شفیرگی و طول عمر حشرات بالغ ماده و نر به ترتیب ۱/۰۸±۵/۸، ۰/۳۳±۰/۶، ۱/۲۱±۸/۳۳ و ۱/۳±۱۲/۶ و ۱/۱±۱۳/۵ روز بدست آمد. میزان باروری ماده‌ها ۱۸/۱±۱۸۲/۳۳ تخم/ ماده محاسبه گردید.