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PHENOLOGY, EMBRYONIC DIAPAUSE AND IMPORTANCE OF NATURAL ENEMIES OF *LEPIDOSAPHES ULMI* (L.) (HOMOPTERA: DIASPIDIDAE) ON OLIVE TREES IN GREECE

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ABSTRACT

Lepidosaphes ulmi (L.) (Diaspididae) develops one annual generation on olive trees in Attica, central Greece. Observations on the phenology of this species were carried out in two localities in Greece, namely Nea Makri (during 1989-1990) and Lavrion (during 1990-1991 and 1991-1992). Eggs began hatching in mid April, after a diapause of more than 8 months. The first males appeared in late May (1990) and in early June (1991). Immature females were observed in early June (1990) and late June (1991), while ovipositing females were present in early July (1990) and mid July (1991). The eggs oviposited by these females in the field entered diapause and hatched, after overwintering, the subsequent April. Eggs collected from Lavrion on July 15, 1991 and transferred to the laboratory (25 ± 1°C, 65 ± 5% RH and 16 hL/8 hD) started hatching 256 days later; however, of the 465 eggs collected, only 2.6% were viable. For 333 eggs, collected from same locality and transferred on January 9, 1992 to the above laboratory regime, the incubation period was 29 days and 66.4% were viable. Eggs collected and transferred to the laboratory on February 26, 1992 hatched within 13 days (99.6% viable), and those collected on March 28, 1992 hatched after 11 days (100% viable). Two predators preyed on this armored scale: *Hemisarcoptes mains* (Shimer) (Acarina: Hemisarcoptidae) being of major importance, and *Cybocephalus fodori* Endrody-Younga (Coleoptera: Nitidulidae) of minor importance. At Lavrion (1991), *H. mains* preyed on live or partly destroyed 2nd-instar females, male pupae, young females, ovipositing females, and on the eggs. At Lavrion, the population of *L. ulmi* decreased 46 times in three months, from 4.6 individuals/cm^2 on May 13, 1991 to 0.1 individuals/cm^2 on August 5, 1991. This reduction is attributed mainly to the predation of *H. mains.* Adults and larvae of *C. fodori* were found in Lavrion in small numbers between May and July 1991.

KEY WORDS: Diaspididae, *Lepidosaphes ulmi*, embryonic diapause, *Hemisarcoptes mains*, olive, Greece.

INTRODUCTION

Originating from Europe and Asia, introduced into America and found in other parts of the world, *Lepidosaphes ulmi* (Linnaeus) (Homoptera: Diaspididae) is a cosmopolitan species able to live in regions with various climates. It infests numerous species of plants of different

families and is distributed all around the Mediterranean basin (Ferris, 1937; Balachowsky, 1954).

In Europe, *L. ulmi* is known as a serious pest of the woody portions of apple trees, as well as other fruit trees and ornamental plants. Among the 25 most frequently recorded species of scale insects, *L. ulmi* was found to be second in order of importance, after *Quadraspidiotus perniciosus* (Comstock) (Kozár and Konstantinova, 1981). In Europe and North America, strains of *L. ulmi* are distinguished on the basis of biological characteristics, such as unisexuality vs. bisexuality, diapause vs. quiescence, number of generations per year, host preference and geographical distribution (Ferris, 1937; Balachowsky, 1954; Gharib, 1978).

On the basis of available information (Alvarado et al., 1975; Argyriou and Kourmadas, 1981; Longo, 1985) the olive-infesting strain of *L. ulmi* in the Mediterranean region could not be clearly distinguished from the strain occurring on apple and other fruit trees in this region. This study was initiated in order to clarify this question and to evaluate the pest status of *L. ulmi* and the role of its natural enemies.

MATERIALS AND METHODS

The identification of *L. ulmi* was confirmed by Danièle Matile-Ferrero, Muséum National d'Histoire Naturelle, Paris, France.

The field work was conducted in two olive groves, on the eastern coast of Attica, central Greece, heavily infested with *L. ulmi,* one at Nea Makri (December 1989–August 1990) and the other at Lavrion about 10 km away (July 1990–April 1992). The grove at Nea Makri contains about 100 old trees (age > 100 years) of the Ladolia variety. The olive grove at Lavrion contains about 100 younger trees (25 years old) of the Manaki variety. In both groves, the trees are 6–8 m apart, in rows. Chemical insecticides were not applied in either grove during the actual study periods.

The phenology of *L. ulmi* was studied at Nea Makri and Lavrion. The spatial distribution of *L. ulmi*, the quantitative estimation of *L. ulmi* infestation level on olive trees and the effect of *L. ulmi* natural enemies were studied only at Lavrion. Samples were taken at 2-week intervals during the warm period of the year and monthly during winter months in both groves. For studying the life cycle of *L. ulmi,* a sample that comprised 20 clearly infested about one-year-old twigs was chosen and cut from different trees (Morgan and Angle, 1969). The twigs were examined in the laboratory and the developmental instars of the scale were recorded.

For monitoring embryonic development, 465 eggs laid during the first half of July were collected on July 15, 1991 at the Lavrion olive grove and transferred to constant conditions (25°C, 65% RH and 16 hL/8 hD) in the laboratory. This process was repeated between January 8 and April 16, 1992, with eggs collected from the field and transferred at 10-day intervals. For checking the stage of embryonic development, eggs were treated and mounted according to the method of Zalokar (1971), as adapted for *L. ulmi* by Gharib and Benassy (1983).

The infestation level of *L. ulmi* was evaluated on samples of four twigs (less than one year old and 20–30 cm long), one from each compass direction at shoulder height, from each of five randomly-chosen trees. The number and developmental stages of live *L. ulmi* individuals were recorded, as well as those dead from unknown causes, attacked by predatory mites and parasitized. The rate of infestation was measured and expressed as number of scales per cm^2 of the plant surface.

Coleopterous predators were sampled by beating four branches on each of five randomly-selected trees with a rubber-covered stick over a 1-m^2 cloth screen, then recording the number of adults and larvae of these predators thus dislodged (Katsoyannos, 1984).

RESULTS

Phenology

Both at Nea Makri (during 1989–1990) and at Lavrion (during 1990–1991 and 1991–1992), *L. ulmi* completed one annual generation (Fig. 1). Its population overwintered at the stage of eggs, protected beneath the scale covers of the females which had laid them. In 1990 crawlers appeared by April 1, and in 1991 by April 18. During March and April 1991, temperatures were lower than in the previous year, which may account for the delay in hatching.

The *L. ulmi* population reproduced bisexually. Male nymphs were found on May 24 in 1990 and on June 3 in 1991. Most female nymphs developed to young, pre-ovipositing adults by June 7 in 1990 and by June 24 in 1991. The majority of these females were ovipositing by

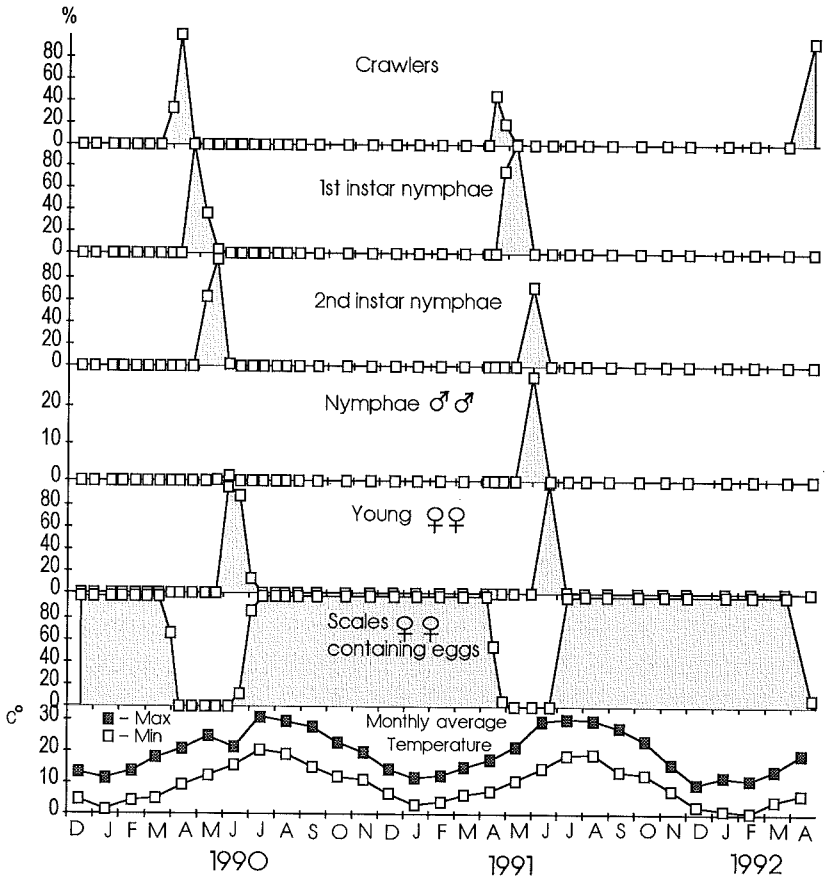

Fig. 1. The life cycle of *Lepidosaphes ulmi* on olive trees at Nea Makri (1989–1990) and Lavrion (1990–1992) in Attica, Central Greece.

July 4, 1990 and by July 15, 1991, respectively. Oviposition lasted for about one month, by the end of which the females had progressively shrunk in body size and died. None of the eggs laid in either year hatched before spring of the following year.

The majority of the *L. ulmi* population was found on woody parts of the olive trees (twigs, as well as older branches and trunks) in both groves. Of 196 *L. ulmi* individuals found on leaves taken from the Lavrion grove during the period June–December 1991, none were alive from July onward (Table 1). No infested fruits were found.

TABLE 1
Number of *Lepidosaphes ulmi* found on leaves of olive trees, Lavrion, Greece

Date	No. of leaves examined	No. of *L. ulmi* individuals							
		1st instar		2nd instar		Male nymphae		Adult females	
		Live	Dead	Live	Dead	Live	Dead	Live	Dead
June 3	223	0	27	15	11	10	8	0	0
June 24	250	0	4	0	6	0	3	0	5
July 7	227	0	3	0	2	0	1	0	9
August 5	195	0	8	0	4	0	1	0	23
August 28	231	0	0	0	4	0	0	0	17
October 7	232	0	7	0	6	0	0	0	8
November 6	185	0	2	0	6	0	0	0	2
December 3	160	0	0	0	2	0	0	0	2
Total	170	0	51	15	41	10	13	0	66

Embryonic diapause

As shown in Fig. 1, *L. ulmi* eggs laid in July 1990 and July 1991 at Lavrion remained beneath the scale covers of the females which had laid them. None hatched in the experiment groves before the following spring.

As shown in Table 2, *L. ulmi* eggs collected from the Lavrion grove on July 15, 1991 and transferred to optimal conditions did not start hatching for many months. The stage of development of the embryos was not checked at the time of collection. A few (8–10) eggs were mounted on slides and checked about 3 months after transfer: these contained embryos with developed thoracic and abdominal segments.

The great majority of eggs collected in July and kept under optimal conditions eventually died. Among the surviving eggs, hatching began after the lapse of more than 256 days and was completed within 21 days, coinciding with the springtime hatching period of *L. ulmi* in the field. The time elapsed from transfer until the first eggs hatched decreased steadily for successive batches of eggs transferred between January 9 and April 17, 1992. By April 17, the number of days elapsed was about one-tenth of the days for the January 9 batch. The duration of the hatching period was about 2 months for eggs transferred between January 9 and February 17.

TABLE 2
Hatching of *Lepidosaphes ulmi* eggs collected from olive trees at Lavrion, Attica, Greece and transferred to optimal (25°C, 65% RH and 16 hL/8 hD) laboratory conditions

Collection dates	Transfer dates	Number of eggs	Number of days elapsed from transfer: First eggs to hatch	>50% of viable eggs hatched	Last eggs to hatch	Viable eggs (%)
1991						
July 15	July 19	465	256	261	277	2.6
1992						
January 8	January 9	333	29	42	84	66.4
January 17	January 18	300	26	38	97	88.0
January 27	January 28	454	21	28	87	93.4
February 5	February 6	1355	15	25	98	98.3
February 17	February 17	1000	15	21	78	99.6
February 26	February 27	1055	13	20	42	99.6
March 7	March 8	1458	15	19	31	99.8
March 17	March 18	953	13	14	23	100
March 28	March 28	153	11	13	20	100
April 6	April 7	497	6	9	27	100
April 16	April 17	961	3	5	24	99.7

Thereafter, the length of the hatching period decreased to less than one month. The rate of viable eggs, representing only two-thirds of the total eggs transferred on January 9, increased to nearly 100% by February 6 and remained high thereafter.

Lepidosaphes ulmi infestation level and its natural enemies

During the period between September 1990 and April 1991 the olive trees at the Lavrion grove were infested with *L. ulmi* at an average density of 2.3 individuals per cm^2 (Fig. 2). On May 13, 1991, *L. ulmi* density peaked at 4.6 individuals per cm^2. Between this date and June 24, *L. ulmi* population density decreased rapidly to 0.4 individuals per cm^2. During July, it further decreased and from August onward, the density was never higher than 0.1 individuals per cm^2.

During the period between May 13 and June 24, the rate of *L. ulmi* found to contain individuals of the predatory mite *Hemisarcoptes malus* (Shimer) (Acarina: Hemisarcoptidae) increased from 0 to 61.4%. *L. ulmi* at different stages of development (2nd-instar nymphs; male nymphs; young, adult pre-ovipositing females; females with eggs) were found containing *H. malus* individuals of different developmental instars. During the same period, the rates of mortality from unknown causes increased more than three times, from 31.9 to 96.0%.

Also found in the Lavrion olive grove at this time were adults of the predator *Cybocephalus fodori* Endrödy-Younga (Coleoptera: Nitidulidae) (10 on May 13 and 3 on June 3, 1991) and *C. fodori* larvae feeding on the bodies of *L. ulmi* nymphs under the scales (6 on June 26 and 3

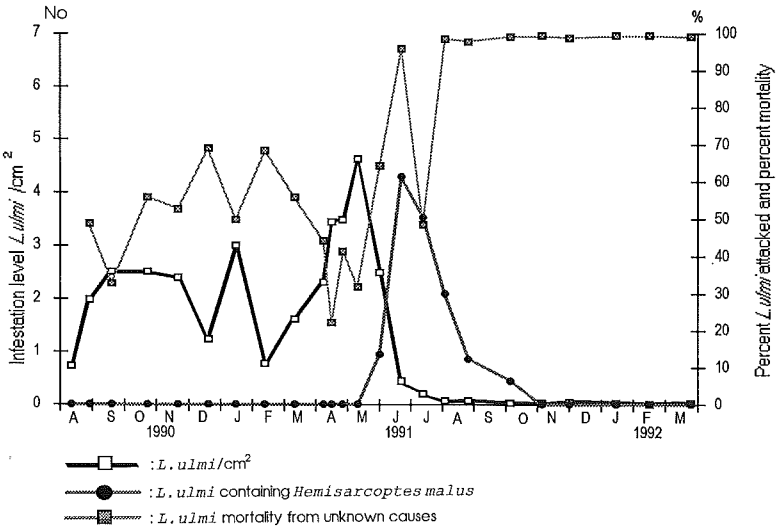

Fig. 2. *Lepidosaphes ulmi* infestation level (individuals/cm^2), *L. ulmi* attacked by *Hemisarcoptes malus* and *L. ulmi* mortality from unknown causes (%) on olive trees at Lavrion, Attica during 1990–1992.

on July 15, 1991). The rate of attack by parasitoids, as estimated by their exit holes, was negligible on *L. ulmi* at both the Nea Makri (<0.2%) and the Lavrion (<0.06%) olive groves.

DISCUSSION

According to Benassy (1986), four strains of *L. ulmi* are distinguished in Europe, based on differences in their mode of reproduction, oviparity or ovoviviparity, annual life cycle, host plants and geographical distribution:

1. A unisexual strain (*L. ulmi unisexualis),* with one generation a year, commonly infesting apple trees in central Europe;

2. A bisexual strain (*L. ulmi bisexualis),* with one generation a year, infesting *Betula* and *Vaccinium* but neither apple nor pear trees in central Europe;

3. A bisexual, oviparous strain without embryonic diapause, developing two or more generations a year on apple, other fruit trees and additional host plants in the Mediterranean region;

4. A bisexual ovoviviparous strain with an obligatory embryonic diapause, developing one generation a year on poplar in the eastern Pyrenees of France. Its full geographical distribution is not yet established (Gharib, 1978).

Based on the evidence of *L. ulmi* with three generations per year on olives in southern Spain (Alvarado et al., 1975), Benassy (1986) concluded that the *L. ulmi* strain which infests Mediterranean olive trees belongs to the third strain mentioned above. This conclusion was also supported by information about *L. ulmi* developing two generations per year on olives in central Greece and southern Italy (Argyriou and Kourmadas, 1981; Longo, 1985).

The findings of the present study suggest that the strain of *L. ulmi* infesting olive trees in Attica, central Greece has more similarities with the fourth strain listed above, the one living on

poplar in the eastern Pyrenees. Indication of embryonic diapause is provided by the phenology results (Fig. 1), showing that eggs laid during July in the field did not hatch before the following spring. Further evidence of the obligatory embryonic diapause of this strain of *L. ulmi* was obtained by transferring newly laid eggs from the field to optimal conditions in the laboratory, in July 1991 (Table 2). These eggs did not start hatching for many months, regardless of the favourable conditions. Eggs of the strain of *L. ulmi* living on poplar in the eastern Pyrenees, collected in July and treated similarly, also did not hatch before January and less than half were viable (Gharib and Benassy, 1983).

Evidence of a progressive termination of the embryonic diapause during the winter months is provided here by the hatching results obtained with eggs transferred to optimal conditions from January 1992 onward (Table 2). These results showed that the time interval between transfer and the first hatching of eggs becomes progressively shorter with each successive batch of eggs: between January and the end of February 1992, the pre-hatching period decreased by half and the percentage of viable eggs increased to its maximum. The same trends were reported for similarly treated eggs of the *L. ulmi* strain living on poplar in the eastern Pyrenees (Gharib and Benassy, 1983).

In view of our findings, it appears that previously reported information about *L. ulmi* infesting olives in the Mediterranean region should be reconsidered. If accidental misidentifications of *L. ulmi* are excluded, the possibility remaining is that more than one *L. ulmi* strain infests the same host plant (olive tree) in the Mediterranean region.

The preference of *L. ulmi* for woody parts of the olive tree, where the great majority of *L. ulmi* in this study were found, agrees with the findings of Argyriou and Kourmadas (1981) and Longo (1985). The high rate of *L. ulmi* mortality recorded on olive leaves at Lavrion (Table 1) indicates that leaves are not a suitable substrate for *L. ulmi*.

The infestation of olive fruits by *L. ulmi* has been reported by Argyriou and Kourmadas (1981) and Longo (1985). In the present study, no infested fruits were found. This may be explained by the fact that the only period of the year in which crawlers were present and able to settle on fruits was April (April 1–26, 1990; April 18–29, 1991) (Fig. 1). Even if olive fruits are susceptible to *L. ulmi*, they were not yet available on the trees, since flowering started in May of both years and the setting of fruits occurred even later.

The doubling of *L. ulmi* infestation level at Lavrion during April and May 1991 (Fig. 2) was due to the hatching of overwintered eggs and the resulting presence of nymphs of the new generation. The drastic decrease in *L. ulmi* population density between May 13 and June 24, 1991, which practically resulted in sanitation of the trees, coincided with a great increase in the proportion of *L. ulmi* individuals attacked by the predatory mite *H. malus*. In *L. ulmi*, there are no distinguishing signs of *H. malus* attack after the event. Therefore, the simultaneous increase of the rate of mortality from unknown causes may be considered at least in part due to the preying activity of *H. malus*. It is likely that the nitidulid predator *C. fodori*, which was also present at Lavrion during late spring–early summer 1991, contributed to the increase in *L. ulmi* mortality rate as well.

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