

## Development and Reproduction of *Lobesia botrana* on Vine and Olive Inflorescences<sup>1</sup>

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### ABSTRACT

Grape vine (*Vitis vinifera* F.) inflorescences were caged in a bag of organdy in the field, and artificially infested with 15 neonate larvae of *Lobesia botrana* (Denis and Schiffenmueller) (Lepidoptera: Tortricidae). Olive twigs, each bearing 5 pairs of inflorescences were caged likewise. In laboratory experiments 15 neonate larvae were placed in each Petri dish containing one vine inflorescence or an olive twig bearing two pairs of inflorescences. Three stages of inflorescence development were used, an early, an intermediate and a late one. Larvae, pupae and adults were maintained at L:D 16:8 and 24°:22°C. In the field, when comparing inflorescences of the earliest developmental stage, the rate of larval development was significantly faster on olive than on vine inflorescences. In the laboratory, when comparing inflorescences of similar developmental stages, the rate of larval development on olive inflorescences was significantly faster than that on vine inflorescences. Pupae of both sexes were significantly heavier on olive inflorescences in all the cases in the laboratory, but in only some cases in the field. In the field the number of eggs per female and the coefficient of multiplication of the insect's population from generation to generation were greater on olive (102.6 and 3.8 respectively) than on vine inflorescences (81.7 and 2.9). In the laboratory, the respective values were 118.5 and 12.3 on olive, and 90.2 and 4.9 on vine inflorescences. In two-choice tests in the laboratory, vine inflorescences were preferred for oviposition to olive inflorescences and to vine or olive leaves. In no-choice tests, vine leaves, vine inflorescences, olive leaves and olive inflorescences in the least advanced stage, were all equally accepted for oviposition.

### Introduction

*Lobesia botrana* is a polyphagous species, with host plants reported to belong to 27 different plant families at least. Vitaceae, Thymeleaceae, Rosaceae, Rhamnaceae, Ranunculaceae, Polygonaceae, Umbelliferae, Compositae, Convolvulaceae and Oleaceae are among them (Balachowsky and Mesnil 1935, Isaakidis 1936, Bovey 1966, Galet 1982, Stoeva 1982). *Daphnae*

*gnidium* (Thymeleaceae) seems to constitute the original host of *L. botrana*. Its adaptation to the grape vine is considered by Balachowsky and Mesnil (1935) to be relatively recent, because at the end of the 19th century the insect was rarely and occasionally found in the vineyards of France and its presence was never generalized as in the early 1930's in that country. Among the Oleaceae reported as hosts of *L. botrana* are the cultivated olive, *Olea europaea* L., and the ornamentals *Syringa vulgaris* L. and *Ligustrum vulgare* L. (Balachowsky and Mesnil 1935,

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Isaakidis 1936, Bovey 1966, Stoeva 1982).

Isaakidis (1936) reports that I. Raftopoulos and N. Mantzoros of the Patras Plant Protection Station in southern Greece, found larvae of *L. botrana* on olive inflorescences. They reared them on olive flowers and obtained adults. Infestations of olive inflorescences by this insect do not seem to be rare, at least in Greece and Bulgaria. Our laboratory stock of the insect was obtained from olive trees near an abandoned vineyard in Halkidiki (Tzanakakis and Savopoulou 1973). E. Angelakis (1987, personal communication) often found larvae on inflorescences of olive trees adjacent to vineyards, on Crete. Stoeva (1982) found in Bulgaria that up to 45% of olive inflorescences were infested with larvae of the first generation. She found pupal length, pupal weight and adult fecundity of field-collected *L. botrana* to be greater on olive inflorescences than on vine inflorescences or sweet cherryfruits.

Recent work in our laboratory (Savopoulou-Soultani and Tzanakakis 1987) showed that on olive inflorescences of the cultivar "Megaritiki" and on vine inflorescences of the cultivar "Razaki" larval growth was fastest in the most advanced stages of inflorescence development. Larval development on olive inflorescences was

approximately 15% faster than on vine inflorescences. By contrast, field experiments showed no significant differences in the speed of larval development between vine and olive inflorescences when we compared inflorescences of similar stages of development. Because of the small number of pupae during that work, pupal weights were not compared and no fecundity and fertility records were kept. Therefore, we considered it advisable to obtain additional data with work on a larger scale, starting with a larger number of larvae and observing, in addition to the duration of larval growth, pupal weight, fecundity, and fertility of adults and to also test the acceptability of olive and vine leaves and flowers as oviposition substrates for *L. botrana*. Such work is reported below.

#### Materials and Methods

The larvae were of our laboratory stock which originated in northern Greece and had been maintained for 16 years on artificial diets we developed (Tzanakakis and Savopoulou 1973, Savopoulou-Soultani and Tzanakakis 1979). The grapevine inflorescences were of the "Razaki" white table cultivar, and the olive inflorescences of the "Megaritiki" cultivar. Three stages of development of

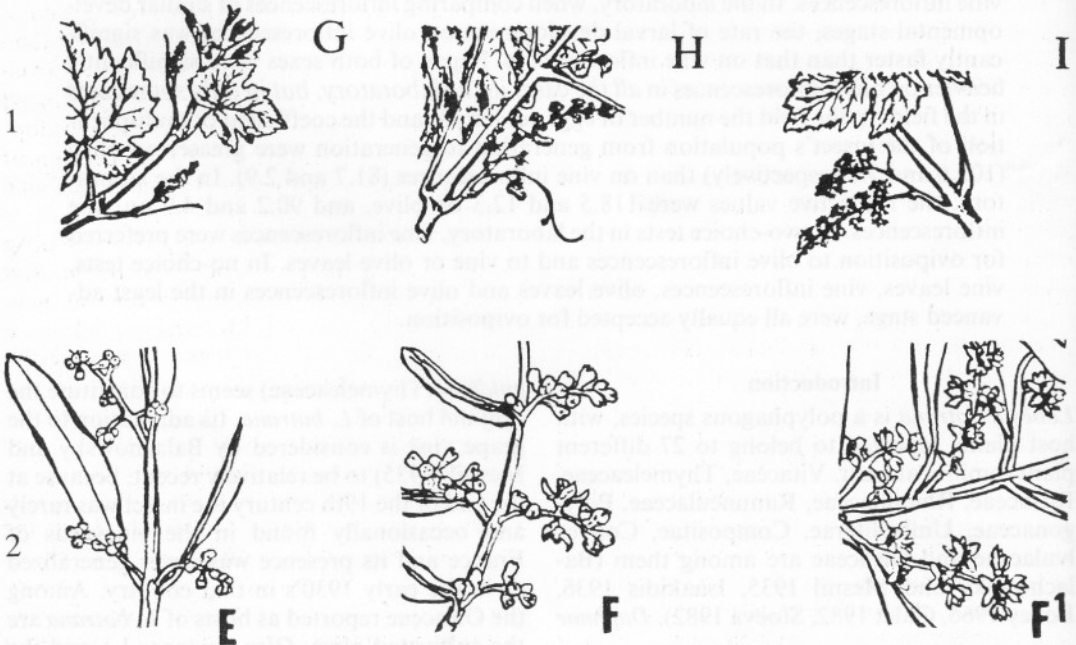


FIG. 1. Stages of development of the vine (1) (after Baggiolini 1967) and of the olive inflorescences (2) (after Colbrant 1981) used in the experiments.

the inflorescences of both plants were compared. For the vine they were G, H and I (Baggiolini 1967) and for the olive E, F, and F<sub>1</sub> (Colbrant and Fabre 1981) (Fig. 1).

In the field, vine inflorescences were used without being removed from the vines in a vineyard at Eginio, 44 km southwest of Thessaloniki and on the University Farm. Fifteen neonate larvae of *L. botrana* were placed on each inflorescence and subsequently caged in a bag of organdy. On the olive trees, the apical part of a twig, bearing 5 pairs of inflorescences was caged likewise, after the leaves from each inflorescence-bearing node were removed and 15 neonate larvae placed on it. There were 7 replicates per treatment. The caged plant parts were checked twice a week, to observe larval development and condition of the inflorescences. Pupation took place in the folds of the organdy bag. Pupae were removed twice a week, taken to the laboratory and maintained at L:D 16:8 and 24°:22°C. They were weighed when 7-10 days old. Of the emerging adults five pairs were maintained at the same conditions in 5 × 7.5 × 9.2 cm truncated conical cups of transparent hard plastic, covered with tissue paper and provided with a cotton wool soaked in 5% sucrose solution, to record fecundity and fertility. The eggs were laid on the walls of the cups.

In the laboratory, one vine inflorescence was placed inside a 9 cm (diameter) glass Petri dish, with a moist piece of cotton at the base of its axis to avoid withering. Likewise, a piece of olive twig bearing 2 pairs of inflorescences was placed in each Petri dish, after the leaves were removed, and a moist piece of cotton was added to its basal end. There were also 7 replicates (dishes) of 15 larvae per treatment. The inflorescences were stored in a refrigerator for up to two weeks, depending on the needs. The vine inflorescences were collected from the University of Thessaloniki Farm, 10 km to the south of the city of Thessaloniki. Larval development and plant part condition were checked daily. Withered or rotten plant parts were replaced promptly with fresh ones taken from cold storage. Rearing took place at L:D 16:8 and 24°:22°C. A piece of corrugated paper, provided the pupation site. Pupae and adults were maintained under the same conditions as those produced in the field.

To determine the population increase from one generation to the next, we calculated the coefficient of population multiplication:  $C = (R/2) \times Lf \times F \times E$  (Guénnelon et al. 1970) where R is the percent of adults per neonate larva, Lf the

percent of mated females, F the number of eggs per female, and E the percent of egg hatchability. R is divided by 2 because the theoretical sex ratio for *L. botrana* is ca. 1.

The oviposition preference experiments were conducted in the laboratory at 16:8 L:D and 25°:23°C. Substrates were inflorescences or leaves. The vine leaves were tender ones, having approximately half their final size. The olive twigs had mostly leaves of the previous year's growing season, followed apically by a few young ones of the current season. All experiments except one were of the choice type. Four females per 15 × 15 × 15 cm cage constituted a replicate. The moths were held in groups of 5 pairs in plastic cups until testing. They were provided with 5% sucrose solution. The females were introduced into the test cages on the next day of their first ovipositions.

Means were compared at the 0.05 level using Duncan's (1955) multiple range test, while percentages were compared using the z-criterion (Steel and Torrie 1960) and oviposition preference using the test for multiple comparisons (Wilcoxon and Wilcox 1964).

## Results and Discussion

### Larval development and survival.

In the field. It is seen in Table 1 that on vine inflorescences which were in stages H and I, larval development was significantly faster than in stage G. On olive inflorescences larval development was also faster in the more advanced stages F and F<sub>1</sub> but not significantly. Development on all three stages of olive inflorescences was slower than on the best two stages of vine inflorescences, but not significantly.

Pupal weights were generally greater on olive inflorescences, than on vine ones, but mostly not significantly so. Pupal survival was generally high on both host inflorescences. In contrast, larval survival was generally low, resulting in a low yield of adults per neonate larva. This low yield in adults could be due to insufficient food for the larvae in each caged inflorescence or twig, or to other factors.

In the laboratory. As seen in Table 1, larval development was significantly faster in the more advanced inflorescences of both plants and slower in the less advanced ones. A comparison of inflorescences of similar stages of development between the two plants, shows that larval development was significantly and substantially faster on olive. Pupal weights were

TABLE 1. Performance of *Lobesia botrana* on vine and olive inflorescences in the field and the laboratory (L:D 16:8, 24°:22°C). (7 × 15 neonate larvae per treatment).

Larval diet	Stage of inflors. development	Date inflors. were caged or picked	Mean duration of larval stage (days)	Mean weight of 7-10 day-old pupae (mg)		Adult as percentage of	
				Males	Females	L1	Pupae
Field experiment							
Vine inflors.	G	24.V.87	30.5a	4.3a	5.7a	11.4a	63.2ac
do.	H	4.VI.87	25.7b	4.7ab	5.8a	12.4a	86.7b
do.	I	7.VI.87	25.2b	4.0a	6.0a	10.5ae	84.6b
Olive inflors.	E	26.V.87	27.7b	4.8ab	6.6ab	27.8c	83.3b
do.	F	7.VI.87	26.4b	5.5b	6.8ab	4.8be	80.0bc
do.	F <sub>1</sub>	12.VI.87	26.0b	5.0b	7.3b	2.9b	66.7ac
Laboratory experiment							
Vine inflors.	G	21.V.87	30.0a	4.2a	7.3a	14.0a	60.0a
do.	H	4.VI.87	24.5b	4.5a	7.9a	17.7b	65.8b
do.	I	10.VI.87	19.0d	4.3a	7.0a	21.9be	59.0a
Olive inflors.	E	29.V.87	22.3c	6.2b	9.7b	21.9ae	76.7bc
do.	F	5.VI.87	17.2d	6.7b	9.3b	33.3d	81.4c
do.	F <sub>1</sub>	10.VI.87	15.5e	6.0b	9.8b	32.4d	77.3bc

Within each experiment and column, numbers followed by the same letter do not differ significantly at the 0.05 level, by Duncan's multiple range test.

also significantly and substantially greater on olive inflorescences. The yield in adults was also significantly greater and pupal mortality lower on olive than on vine inflorescences. Therefore, in the laboratory, olive inflorescences were superior to vine ones as food for larvae of *L. botrana*.

#### Fecundity and fertility

As seen in Table 2, the number of eggs per female and the coefficient of multiplication of the insect's population from generation to

generation were greater on olive than on vine inflorescences, in both field- and laboratory-reared larvae. Egg hatchability was not significantly different between the two plants in either field- or laboratory-reared insects.

#### Oviposition preference

As seen in Table 3, in the two-choice tests vine inflorescences were preferred for oviposition to olive inflorescences and to vine or olive leaves. Vine leaves were preferred to olive inflorescences in stage F but not in stage E.

TABLE 2. Reproduction at L:D 16:8 and 24°:22°C of *Lobesia botrana* reared as larvae on vine and olive inflorescences in the field and in the laboratory.

Larval diet	No. of females	Adults as percentage of L <sub>1</sub>	Mated females (%)	Mean no. of eggs/female	Egg hatchability (%)	Coefficient of multiplication from generation to generation
Reared in the field						
Vine inflors.	20	11.4	88.9a	81.7a	69.1a	2.9
Olive inflors.	25	11.8	94.3a	102.6b	65.8a	3.8
Reared in the laboratory (L:D 16:8, 24°:22°C)						
Vine inflors.	30	21.2	85.3a	90.2a	72.6a	4.9
Olive inflors.	40	29.2	90.1a	118.5b	78.9a	12.3

Within each experiment and column, numbers followed by the same letter do not differ significantly at the 0.05 level, by Duncan's multiple range test.



TABLE 3. Oviposition preference of *Lobesia botrana* on flowers and leaves of vine and olive in two-choice and no-choice tests.

Oviposition substrate	No. of replicates	Stage of inflors. development	Mean no. eggs/replicate/day (4♀/replicate)
Two-choice tests			
Vine inflorescences	11	G	15.0a
Olive inflorescences		E	0.0b
Vine inflorescences	12	G	32.0a
Vine leaves			0.0b
Vine inflorescences	12	G	9.0a
Olive leaves			3.0b
Vine leaves	4		3.0a
Olive inflorescences		E	2.0a
Vine leaves	11		18.0a
Olive inflorescences		F	8.0b
Vine leaves	10		10.5a
Olive leaves			5.0b
Olive leaves	5		2.0a
Olive inflorescences		E	1.0a
Olive leaves	11		28.0a
Olive inflorescences		F	4.0b
No-choice test			
Vine inflorescences	7	G	18.0a
Vine leaves	8		17.5a
Olive inflorescences	5	E	8.0ab
Olive inflorescences	8	F	2.0b
Olive leaves	7		12.0a

Within each test, means followed by the same letter do not differ significantly at the 0.05 level, by Wilcoxon and Wilcoxon test for multiple comparisons.

They were also preferred to olive leaves. Olive leaves were preferred to olive inflorescences in stage F but not in stage E. As seen in the no-choice test, significantly and substantially fewer eggs were laid on olive inflorescences when in stage F, than on vine inflorescences, or on vine and olive leaves. Therefore, when the moths had no choice, they accepted vine leaves, olive leaves and olive inflorescences in stage E equally well as vine inflorescences.

The present work substantiates the previous year's preliminary results, that larvae developed on olive inflorescences as fast and in some cases faster than on vine inflorescences and that the pupae were heavier. It also proves that, both in the field and in the laboratory, the adults produced on olive inflorescences give more eggs

than those produced on vine inflorescences. Yet, the female moths, in the absence of vine inflorescences, laid as many eggs on olive leaves and inflorescences (stage E) as on vine leaves and inflorescences. In view of the fact that *L. botrana* oviposits readily on olive inflorescences and that an ordinary olive grove adjoining a vineyard may, in most years, offer an abundance of olive flowers, there is no question that an olive grove may contribute to the production of large numbers of first-generation adults of this insect, which may lay more eggs than those developed on vine. Therefore, olive trees may constitute an important source of infestation of nearby vines by moths of the first generation. Such a source of infestation should be taken in consideration when planning control measures against *L. botrana*.

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KEY WORDS: Insecta, Tortricidae, *Lobesia botrana*, Host-plant suitability, Oviposition preference, Development, Fecundity, Grapevine pests, Olive insects

## Ανάπτυξη και Αναπαραγωγή του *Lobesia botrana* σε Ανθοταξίες Αμπέλου και Ελιάς

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### ΠΕΡΙΛΗΨΗ

Έγιναν πειράματα τόσο στην ύπαιθρο όσο και στο εργαστήριο, με σκοπό να διευκρινιστεί ο ρόλος της ελιάς ως ξενιστή του *Lobesia botrana* (Dennis και Schiffermueller) σε σχέση με την άμπελο. Στα πειράματα υπαίθρου παρατηρήθηκε η ανάπτυξη της προνύμφης σε ανθοταξίες αμπέλου και ελιάς σε 3 διαφορετικά στάδια ανάπτυξης της ανθοταξίας, πρώιμο, ενδιάμεσο, αναπτυγμένο. Για το σκοπό αυτό εγκλωβίστηκαν 7 ανθοταξίες αμπέλου σε σάκκους από οργαντίνα αφού προηγουμένως τοποθετήθηκαν 15 νεοεκκολαφθείσες προνύμφες σε κάθε ανθοταξία. Στην ελιά εγκλωβίστηκαν με τον ίδιο τρόπο κλαδίσκοι που έφεραν 5 ζεύγη ανθοταξιών, αφού αφαιρέθηκαν τα φύλλα από τους κόμβους που έφεραν τις ανθοταξίες. Δύο φορές την εβδομάδα γινόταν

έλεγχος της εξέλιξης των προνυμφών και της κατάστασης των ανθοταξιών. Οι προνύμφες νυμφώνονταν στις πτυχές του σάκκου. Οι νύμφες μαζεύονταν 2 φορές την εβδομάδα και μεταφέρονταν στο εργαστήριο όπου διατηρούνταν σε χώρο με φωτοπερίοδο L:D 16:8 ωρών και θερμοκρασία 24°:22°C. Ζυγίζονταν σε ηλικία 7-10 ημερών. Τα ενήλικα τοποθετούνταν ανά 5 ζεύγη σε πλαστικά διαφανή κύπελλα με διαστάσεις 5 × 7.5 × 9.2 cm σκεπασμένα με χαρτοπετσέτα. Για τροφή τους είχαν βαμβάκι εμποτισμένο με διάλυμα ζάχαρης 5%. Στα πειράματα εργαστηρίου τοποθετήθηκε μέσα σε τρυβλίο Petri μία ανθοταξία αμπέλου ή ένας βλαστός ελιάς με 2 ζεύγη ανθοταξιών και 15 νεοεκκολαφθείσες προνύμφες. Οι ανθοταξίες αντικαθιστούνταν όταν μαραίνονταν. Και στην περίπτωση αυτή χρησιμοποιήθηκαν 3 στάδια ανάπτυξης των ανθοταξιών. Η εκτροφή του εντόμου έγινε στις πιο πάνω συνθήκες.

Τα πειράματα προτίμησης φωτοκίας έγιναν στο εργαστήριο σε L:D 16:8 και 25°:23°C, σε μικρά μεταλλικά κλουβιά, που στο καθένα τοποθετήθηκαν 4 θηλυκά. Ως υποστρώματα φωτοκίας τοποθετούνταν ανθοταξίες και φύλλα ελιάς ή αμπέλου. Όλα τα πειράματα εκτός από ένα ήταν του τύπου διπλής επιλογής.

Τα πειράματα υπαίθρου έδειξαν ότι η ανάπτυξη των προνυμφών στα 3 διαφορετικά στάδια εξέλιξης των ανθοταξιών στην ελιά ήταν βραδύτερη από εκείνη στα στάδια Η και Ι της αμπέλου, αλλά όχι σημαντικά, αλλά σημαντικά ταχύτερη από του σταδίου G. Οι νύμφες που προήλθαν από άνθη ελιάς ήταν βαρύτερες, χωρίς η διαφορά να είναι σε όλες τις περιπτώσεις στατιστικά σημαντική. Η νυμφική θνησιμότητα ήταν χαμηλή σ' όλες τις περιπτώσεις, ενώ αντίθετα, η προνυμφική θνησιμότητα ήταν υψηλή.

Τα πειράματα εργαστηρίου έδειξαν ότι η ανάπτυξη της προνύμφης ήταν σημαντικά ταχύτερη στις ανθοταξίες της ελιάς απ' ότι της αμπέλου σε αντίστοιχα στάδια εξέλιξης των ανθοταξιών. Οι νύμφες από ανθοταξίες ελιάς ήταν σημαντικά βαρύτερες από εκείνες που προήλθαν από ανθοταξίες αμπέλου σ' όλες τις περιπτώσεις. Η απόδοση σε ενήλικα ήταν μεγαλύτερη και η νυμφική θνησιμότητα μικρότερη στις ανθοταξίες ελιάς απ' ό,τι στις αμπέλου.

Ο αριθμός αυγών ανά θηλυκό και ο συντελεστής αυξήσεως του πληθυσμού από γενεά σε γενεά ήταν μεγαλύτεροι στα άτομα που αναπτύχθηκαν σε ανθοταξίες ελιάς απ' ό,τι σ' εκείνα που αναπτύχθηκαν σε ανθοταξίες αμπέλου, τόσο στην υπαίθρο όσο και στο εργαστήριο.

Όσον αφορά την προτίμηση φωτοκίας, τα πειράματα διπλής επιλογής έδειξαν ότι οι ανθοταξίες αμπέλου προτιμούνται ως υπόστρωμα φωτοκίας από εκείνες της ελιάς και από τα φύλλα ελιάς ή αμπέλου. Τα φύλλα αμπέλου προτιμούνται από τα φύλλα και τα άνθη ελιάς σταδίου F αλλά όχι από εκείνα σταδίου E. Τα φύλλα ελιάς προτιμούνται από τα άνθη σταδίου F αλλά όχι από εκείνα σταδίου E. Στο πείραμα χωρίς επιλογή γεννήθηκαν σημαντικά λιγότερα αυγά σε ανθοταξίες ελιάς στο στάδιο F απ' ό,τι στο στάδιο E, ή σε ανθοταξίες αμπέλου ή σε φύλλα ελιάς ή αμπέλου.

Συμπεραίνουμε ότι τα φύλλα και οι ανθοταξίες ελιάς, σε ορισμένα στάδια ανάπτυξης, απουσία ανθοταξιών αμπέλου, γίνονται δεκτά ως υπόστρωμα φωτοκίας του *L. botrana*. Οι προνύμφες αναπτύσσονται εξίσου γρήγορα και υπό ορισμένες συνθήκες γρηγορότερα σε άνθη ελιάς και δίνουν ενήλικα που έχουν υψηλή ωσπαραγωγική ικανότητα. Συνεπώς, ελαιώνες που βρίσκονται κοντά σε αμπέλια μπορεί να αποτελέσουν εστίες παραγωγής αξιόλογων πληθυσμών του εντόμου στην πρώτη γενεά. Αυτό πρέπει να λαμβάνεται σοβαρά υπόψη όταν προγραμματίζονται μέτρα καταπολέμησης του εντόμου αυτού.