Differences in nutritional quality of parts of *Vitis vinifera* berries affect fitness of the European grapevine moth

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Abstract

The European grapevine moth, *Lobesia botrana* (Denis & Schiffermüller) (Lepidoptera: Tortricidae), is a major grapevine pest in Europe. The larva is polyphagous and able to develop on more than 25 plant species, several of them being more suitable than Vitaceae for the fitness of *L. botrana*. Larvae normally eat the pulp of the berry, but may also consume the seeds according to the development stage of the berry and the larval density per bunch. Understanding the effect on individual fitness of such feeding behaviour is important to assess how suitable the different berry tissues are for this insect. We offered to the larvae either entire berries, seeds, or pulp with skin of the variety *Vitis vinifera* cv. 'Cabernet Sauvignon' as larval food in order to assess several life history traits from egg hatching to adult death. Two control groups were raised on semisynthetic diets, offering a low (no plant material) or a higher (with corn flour) nutritive value medium. The larvae performed differently when fed on diets containing different parts of berries. The larvae fed on a diet containing grape berry seeds had a prolonged development time and showed higher mortality. The females emerging from these larvae had a lower fecundity and mating success than the females emerging from larvae fed on diets containing other parts of the berries. However, their longevity was greater in comparison to the other groups. We conclude that seeds of Cabernet Sauvignon are not toxic to larvae but affect the reproductive life history traits in *L. botrana*.

Introduction

The European grapevine moth (EGVM), *Lobesia botrana* (Denis & Schiffermüller) (Lepidoptera: Tortricidae), is a major grapevine pest in the palearctic region (Bovey, 1966; Roehrich & Boller, 1991). Despite the abundance of vineyards across Europe, this insect is still a generalist; females are attracted and able to oviposit on alternative plants (Maher et al., 2000; Thiéry, 2005; Thiéry & Moreau, 2005), and larvae are able to complete their development on more than 25 plant species belonging to various families. (Balachowsky & Mesnil, 1935; Bovey, 1966; Stoeva, 1982; Savopoulou-Soultani et al., 1990; Thiéry, 2005).

Several studies have shown that among the alternative host plants, flowers of the olive, *Olea europaea* L. (Oleaceae), or berries of the flax-leaved daphne, *Daphne gnidium* L. (Thymeleaceae), for example, are more suitable than Vitacea for the performance of *L. botrana*: larval mortality and development time of the larvae are reduced, whereas pupal weight, female longevity, female fecundity, and mating success are increased (Stoeva, 1982; Savopoulou-Soultani et al., 1990; Stavridis & Savopoulou-Soultani, 1998; Thiéry & Moreau, 2005). One of the major questions raised by these studies is why grapes are less suitable for EGVM larval development than other plants.

For insects that do not feed during the adult stage, the quantity and quality of food ingested as larvae strongly influence the amount of reserves stored in the abdominal fat body that is allocated for reproduction during the adult phase (Slansky & Scriber, 1985; Slansky & Rodriguez,

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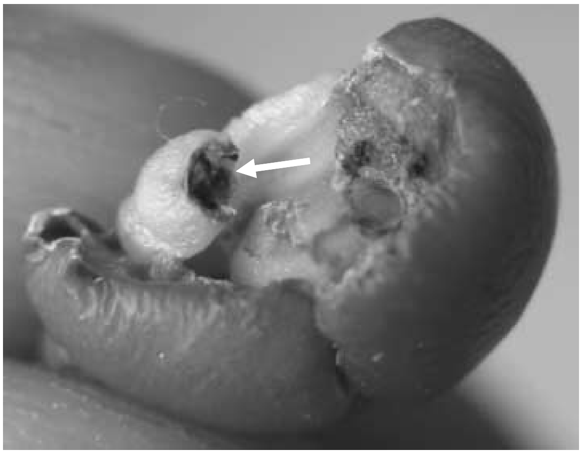

Figure 1 Seed of Cabernet Sauvignon (stage 32) eaten by a *Lobesia botrana* larva (white arrow).

1987; Awmack & Leather, 2002). Given the importance of larval food quality, one possibility to explain why grapes are less suitable for *L. botrana* than other host plants is that grape berries have a lower nutritive value than fruits of other plants. In this context, the presence of seeds in the berries of grapes may play an important role. Under natural conditions, larvae normally avoid eating the seeds. However, during the second adult generation (at the beginning of July in France), larvae can eat them when the amount of pulp is limited and when the seeds are not yet too hard (Figure 1 and D Thiéry, unpubl.). This depends also on the larval density per bunch (D Thiéry, unpubl.). At veraison, seeds become harder and richer in polyphenols such as flavonol monomers, which occur concentrated in the seed coat and were shown to increase five-fold in a Shiraz cultivar (Kennedy et al., 2000). The larvae avoid ingesting the seeds at this stage, but whether the reason for this avoidance is chemical or physical has not been elucidated yet.

The present experiments were designed to evaluate the nutritional consequences of eating the different parts of the berry for the EGVM larva, when larvae may eat seeds during the second adult generation. We hypothesized that the presence of seeds in the berries may reduce larval and adult performance when larvae eat them, as their polyphenol concentration is relatively high (Kennedy et al., 2000; Peng et al., 2001; Geny et al., 2003), and mainly because tannins are known to reduce the digestibility of proteins (Winkler, 1962) and may have a toxic or feeding deterrent effect on phytophagous insects (Hegarty et al., 1991). This hypothesis was assayed on diets supplemented with berries or parts of berries harvested before berries change colour. We thus tested the effect of seeds alone and pulp with skin on several life history traits of *L. botrana*. We compared these results to those obtained with entire berries and two artificial diets of high and low nutritional quality. The effect of different diets on larvae and adults was determined by measuring the complete suite of life history traits of *L. botrana*, from egg hatching to adult death. Particularly, we recorded several developmental life history traits such as pupal mass, larval development time, and larval survival. We also determined the influence of diet on different components of female reproductive success such as mating success, fecundity, and female longevity.

Materials and methods

Insect origin and maintenance

We used a strain of *L. botrana* (INRA Bordeaux, France) originating from individuals collected in a French Sauternes vineyard (cultivar white sauvignon) and maintained as a stock colony for 10 generations on a semi-artificial diet described in Mondy (1998) at 24 ± 1 °C, 60 ± 10% r.h., with a photoperiod of L15:D8 and 1 h of dusk. The photophase was provided by daylight fluorescent tubes (15 h at 1000 lux luminosity and 1 h equivalent to dusk at 25 lux). All tests were performed under these standard conditions.

Characteristics of grapes and berries

We used *V. vinifera* cv. ‘Cabernet Sauvignon’ from our experimental vineyard ‘Domaine de la Grande Ferrade’, INRA Bordeaux. The vineyard was planted in 1973 with eight rows of 35 stocks. Rows and stocks were spaced 1.2 and 1.1 m, respectively, and no fertiliser, fungicides, or insecticides were used during the year preceding the experiments. Berries were harvested before veraison corresponding to phenological stage 34–35 (Baillod & Baggiolini, 1993), which occurs in mid-July in the Bordeaux region. This development stage of the grapes is infested by the second annual generation of *L. botrana*. Berries were hard and green and seeds were plump, bright green, and pliable. Bunches were picked and taken to the lab, where all the berries were cut lengthwise. In a part of them, the seeds were removed from the rest of the berry. All the material was immediately stored at –18 °C before use.

Larval diets and general procedure

Because grape seeds and berries without seeds cannot be tested in their natural state, their effect was evaluated using a standardized procedure similar to that described elsewhere (Thiéry & Moreau, 2005). It is based on an artificial diet for Lepidoptera in which freeze-dried powder of plant material is incorporated. Berries were freeze-dried at –54 °C, under 140 mbar during 24 h (Christ alpha 1–4, Fischer, Bioblock Scientifics, Illkirch, France), and a fine powder was obtained using a stainless steel blender (Waring, New Hartford, CT, USA) at 10^4 r.p.m. for 2 min.

This protocol was used because it allows one to test the seeds and the rest of the berry separately. It also has the advantage of offering equivalent amounts of food to the larvae and to prevent biases due to competition and subsequent food deprivation as feeding larvae are raised separately. It also avoids the occurrence of fungi such as *Botrytis cinerea*, which grow naturally on berries and which may modify larval fitness (Savopoulou-Soultani & Tzanakakis, 1988; Mondy & Corio-Costet, 2000).

Larvae were raised individually to pupation in Eppendorf tubes filled with 1.5 ml of an agar-based medium composed of (for 100 Eppendorfs): 150 ml water, 5 g agar, 6 g cellulose powder, 4 g vitamin-free casein, 3.5 g glucose, 2 g mineral salt, 0.12 g cholesterol, 0.12 g maize oil, 0.25 g benzoic acid, 0.1 g nipagine, and 12 g freeze-dried powder of (1) seeds, (2) pulp and skin, or (3) entire berries. Chemicals used were purchased from ICN biochemicals (Irvine, CA, USA), except maize oil (commercial source, cooking quality).

A control group of larvae was reared on this medium with no plant material (called Basic Medium, BM) corresponding to a medium with low nutritive value (Thiéry & Moreau, 2005). A complementary larval rearing programme was conducted on an artificial diet [called Augmented Medium (AM)], supplemented with non-host plant material but with some elements with high nutritive value (corn flour, wheat germ, and yeast) under the rearing conditions described previously (Thiéry & Moreau, 2005). Its composition was as follows (source as specified previously unless otherwise stated): 150 ml water, 3 g agar, 9 g maize flour, 11 g wheat germ, 9 g yeast (INRA, Le Magneraud, France), 0.9 g ascorbic acid (Sigma Aldrich, St Louis, MN, USA), 0.3 g benzoic acid, 0.3 ml maize oil, 0.3 g methyl-p-hydroxybenzoate, nipagine, used to prevent *Nosema* disease, and 0.2 g iprodione (Rovral, Bayer Crop Science, Lyon, France) used as fungicide. These two kinds of artificial diets are classically used for rearing *L. botrana* for successive generations, and all the chemicals listed previously were previously checked for lack of toxicity on the larvae (D Thiéry, unpubl.).

All Eppendorf tubes were pierced to allow air circulation. Newly hatched larvae (age <24 h) were transferred individually using a fine brush to an Eppendorf tube. One hundred larvae were assigned to each treatment, divided in two batches.

Developmental time and pupal weight

The first batch of 40 larvae on each diet was monitored daily until pupation. Pupae were taken from the diet and weighed to the nearest 0.1 mg as an index of the adult body size. Pupae were placed individually in glass tubes (70 mm long × 9 mm in diameter) covered with a cotton plug and stored in the test room until emergence. Adults were sexed upon emergence. We recorded the following variables: (1) total larval development time for each sex, defined here as the number of days from egg hatching until adult emergence; (2) global larval survival corresponding to the proportion of emerged moths; (3) pupal mass of each sex; and (4) adult sex ratio.

Reproductive life history traits

The second batch of 60 larvae per dietary treatment (not monitored daily) was only used to produce additional adults needed to test the reproductive output of females. Therefore, all emerged adults from the two batches were used to evaluate the reproductive output of females. Newly (less than 1-day-old) emerged females were confined individually in 0.5-l cellophane bags as mating and oviposition chambers and provided with water ad libitum through a soaked cotton dental wick. One- or 2-day-old virgin males originating from the same diet were added to each caged virgin female, 1 h before dusk. Males were randomly assigned to females. Pairs were caged in these bags until the death of both sexes. Females oviposited freely inside the cellophane bag until their death.

We considered four variables: (1) percentage of mated females (female mating success) assessed by production of at least five fertile eggs during her lifetime (non-mated females are able to lay some infertile eggs at the end of their life), (2) fecundity (mean number of eggs laid per female), (3) oviposition period (corresponding to the duration of the egg laying period), and (4) longevity of mated females.

Statistical analysis

All statistics were performed using JMP software (version 3.2.2, SAS Institute, 1995). A preliminary stepwise analysis (backward procedure) was used to remove non-significant ($P>0.05$) effects and interactions on life history traits assessed (Quinn & Keough, 2002). Only the resulting models are presented here. Therefore, to detect the effect of diet on percentage of larval + pupal mortality, female ratio, and percentage of mated female, we used a Pearson χ^2 test (Sokal & Rohlf, 2001). To detect the effect of diet on male and female development time, fecundity, oviposition period, and longevity of females, we used a Kruskal–Wallis test, as data did not meet normality (Shapiro–Wilk test) or homogeneity of variances assumptions (Levene test) (Sokal & Rohlf, 2001). Two-tailed tests of significance were used throughout.

Results

Effect of larval diet on growth and development

Development times of males and females were significantly different according to the diet on which larvae were fed

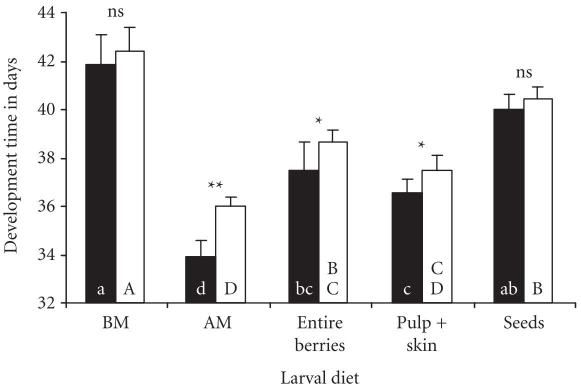

Figure 2 Development time in days (mean + SEM) of *Lobesia botrana* females (white bars) and males (black bars) reared on five larval diets (BM, basic medium; AM, augmented medium). Bars with the same letters indicate non-significant differences (P>0.05) after a non-parametric PLSD test (capital letters for females and small letters for males). Comparisons between males and females was done with a Kruskal–Wallis test; ns, P>0.05; * P<0.05; ** P<0.01.

(Figure 2, Kruskal–Wallis test: for females: $\chi^2 = 30.36$, $P<0.0001$; and for males: $\chi^2 = 36.01$, d.f. = 4, $P<0.0001$). The AM diet resulted in the shortest larval development times, whereas BM produced the longest. Inclusion of seeds led to a longer female larval development than pulp alone, whereas entire berries did not significantly differ from seeds alone or pulp + skin alone. In three of the diets that promoted fast larval development (AM, entire berries, and pulp + skin), males emerged sooner than females, whereas in the two others that produced slow larval development (BM and seeds), the time difference was not significant (Figure 2).

Pupal mass varied significantly according to diet and sex (females were heavier than males) (Figure 3, Standard Least Squares: general model: $F_{5,129} = 34.46$, $P<0.0001$; effect of diet: $F_{4,129} = 8.29$, $P<0.0001$; effect of sex: $F_{1,129} = 263.79$, $P<0.0001$). The significant difference between diets is mainly due to the BM, as larvae that fed on BM reached a lower weight than those fed on the other diets. However, although there was a difference in the larval development time when the larvae were reared on the diets containing different parts of the berry, the mass of pupae (female and male) was not different.

The percentage of larval + pupal mortality differed between diets (Table 1); the lowest percentages of mortality were observed on the AM diet. Larvae fed on entire berries exhibited a lower mortality than those fed on pulp + skin only; larvae fed on seeds exhibited the highest rate of larval mortality. There was no effect of diet on sex ratio of the emerging adults.

Effect of larval diet on reproduction

Female mating success, measured as the number of females that laid at least five fertile eggs, ranged from 68 to 91% and was significantly different between diets (Table 1). In particular, females that were raised on seeds and BM were less successful at mating than those raised on AM, entire berries, and pulp + skin. Lifetime egg production strongly varied depending on diet. Females were least fecund on BM and seeds although females from AM and pulp + skin exhibited the highest fecundity (Table 1). Females raised on entire berries exhibited a fecundity intermediate between those raised on seeds and pulp + skin. This variation in fecundity is partially explained by the duration of egg laying during the life. Females raised on AM laid eggs for a longer time than those raised on BM and entire berries (Table 1). Although females raised on seeds have a lower fecundity, they laid for a longer period. Finally, the longevity of females varied depending on larval diet (Table 1). Females raised on seeds lived longer than other females and even than those raised on AM.

Discussion

Our results support the hypothesis that quality of larval food is decisive for *L. botrana* larval duration and adult performance. The results show that life history traits of *L. botrana* vary according to the type of food consumed in the larval stage: (1) newly emerged females that fed on seeds during the larval stage had a prolonged larval development, lower fecundity, and lower mating success than females that fed on other parts of berries; (2) the

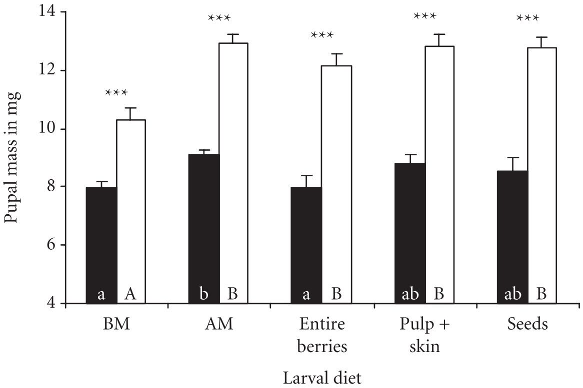

Figure 3 Pupal mass (mean + SEM) of male (black bars) and female (white bars) *Lobesia botrana* reared on five larval diets (BM, basic medium; AM, augmented medium). Bars with the same letters indicate non-significant differences (P>0.05) after a non-parametric PLSD test (capital letters for females or small letters for males). Comparison between males and females was done with a Kruskal–Wallis test, *** P<0.0001.

Table 1 Performance of *Lobesia botrana* according to the diet on which the larvae developed. Values (mean ± SEM) in each column with the same letter are not significantly different (P>0.05) after a non-parametric PLSD test

	Growth and development		Reproductive life history traits			
Larval diet	Percent larval + pupal mortality	Female ratio (%)	Percent of mated females (number of female tested)	Mean number of eggs laid per female	Oviposition period in days	Longevity of female in days
Basic medium	29.03	45.45	69.57 (23)	83.37 ± 5.29d	3.75 ± 0.40e	8.75 ± 0.81c
Augmented medium	8.82	38.71	88.46 (26)	196.17 ± 6.53a	8.13 ± 0.42a	10.83 ± 0.41c
Entire berries	28.20	42.85	87.88 (33)	122.28 ± 6.64c	4.79 ± 0.27d	7.00 ± 0.31d
Pulp + skin	35.90	48.00	90.91 (33)	144.77 ± 7.53b	6.03 ± 0.36c	8.53 ± 0.37c
Seeds	40.00	45.83	68.09 (47)	96.69 ± 10.46d	7.12 ± 0.22b	13.50 ± 0.31a
χ^2 (d.f. = 4)	9.99[1]	0.58[1]	10.73[1]	58.75[2]	55.46[2]	75.58[2]
P	0.04	0.97	0.03	<0.001	<0.0001	<0.0001

[1] Pearson χ^2 test; [2] Kruskal–Wallis test.

mortality of larvae fed on seeds was highest; (3) on the other hand, the longevity of females raised on seeds was strongly prolonged in comparison to others. This phenomenon was not observed for insects raised on entire berry medium.

Larval development time varied among diets as demonstrated before (Savopoulou-Soultani et al., 1999; Thiéry & Moreau, 2005). Basic Medium and seed diets induced longer larval development in comparison to the other diets. The similarity between BM and the seed diet could be explained by either a low nutritive value or by a lack of phagostimulants in these two diets. However, because the seed diet produced pupae of similar weight to those obtained with the other diets, we may consider that the amount of food ingested was similar and thus the differences observed can be attributed to food quality only.

Three diets supported a fast larval development (AM, entire berries, and pulp + skin), and the emergence of males occurred before females. However, in the two diets promoting a slow development (BM and seeds), this delay was not significant. Protandry occurs in many Lepidoptera including *L. botrana* (Thomas, 1989; Wiklund et al., 1993; Savopoulou-Soultani et al., 1994; Torres-Vila et al., 1995) and presents some advantages by maximizing copulation opportunities for a male (reviewed in Wiklund & Fagerström, 1977; Bulmer, 1983), and by minimizing the prereproductive period of females as they emerge when most males are available (Fagerström & Wiklund, 1982). Our results indicate that this protandry depends upon the nutritional value of the diet.

Furthermore, females that were raised on the seed diet had higher mortality than larvae fed on entire berries or pulp + skin. This supports the hypothesis that seeds represent a diet of poor quality for *L. botrana* larvae. However, although larvae fed on seeds exhibited the highest larval and pupal mortality, larvae fed on entire berries exhibited lower larval mortality than larvae fed on pulp + skin. This means that entire berries probably offer to the larvae a more complete set of nutrients produced by each part of the berry (seeds and pulp + skin) necessary for good survival.

Interestingly, although larval development time clearly varied between diets, the resulting pupae had similar weights, except for those raised on medium with very low nutritive value (BM). The positive relationship between size and development time appears in general to be less strict in butterflies than in some other animals due to plasticity in growth rate (Wiklund et al., 1991). Here, prolonged larval development time on the seed diet enabled larvae to compensate for food of poor quality and finally to reach nearly the same weight as larvae that were reared on diets that promoted fast larval development. Usually, females of *L. botrana* are heavier and larger than males, a trait that is mainly attributed to the storage of nutrients used for egg production (Raven, 1961; Slansky & Scriber, 1985). Female larvae certainly achieve a greater size by feeding and developing for a longer period of time (Mackey, 1978; Lederhouse et al., 1982) as observed during our experiments.

The results reported here also confirm the importance of larval food for adult reproductive success (Torres-Vila et al., 1999; Torres-Vila & Rodriguez-Molina, 2002; Thiéry & Moreau, 2005) as it influenced the mating success of the resulting adult confirming earlier results (Thiéry & Moreau, 2005; Moreau et al., 2006). Indeed, a decrease in female mating success occurred after development on seeds and BM. The reason for this remains to be explained, but it is now well known that host plants (or diets) may play an important role in the production and release of pheromones in some phytophagous insects (including moths) through the acquisition of chemical precursors of pheromones from plant material (Landolt & Phillips, 1997). Here, we cannot exclude that compounds required for the production of pheromones are missing in several diets, resulting in low mating frequency.

Larval diet also strongly influences female fecundity as demonstrated before in this species (Torres-Vila et al., 1999; Thiéry & Moreau, 2005). Females obtained from the BM and seed diets exhibited low fecundity (less than 100 eggs laid), whereas those from the AM, entire berries, and pulp + skin diets exhibited high fecundity (above 130 eggs). These results indicate that a high food quality promotes faster larval development and high fecundity, which suggests that no fitness penalty is paid for fast development. Despite the disadvantages resulting from feeding on seeds, the longevity of the resulting females was greater and this may have consequences on their dispersal capacity and also the capability of successive mating. This was, however, not tested here.

In conclusion, seeds are less suitable than the pulp + skin or the entire berry for almost all life history traits of *L. botrana* (prolonged larval development, high mortality and low mating success, and fecundity). The presence of seeds in grape berries could explain why the berries of grapes are less suitable food than berries of alternative host plants as demonstrated by several authors (Stoeva, 1982; Savopoulou-Soultani et al., 1990; Stavridis & Savopoulou-Soultani, 1998; Thiéry & Moreau, 2005). These differences in suitability between different parts of berries may be explained by differences in nutritive quality but also by secondary compounds, e.g., tannins that reduce protein digestibility or act as feeding deterrents (Winkler, 1962; Hegarty et al., 1991). Recently, high levels of tannins were found in seeds of grape berries (*V. vinifera* cv. Shiraz) (from 1360 to 2830 mg kg^{-1} of berries; Peng et al., 2001; Geny et al., 2003), and the seeds of 'Cabernet Sauvignon' berries exhibited the highest contents of tannins before ripening in comparison to other cultivars (Harbertson et al., 2002). Our results lead us to conclude that seeds impair the individual fitness of *L. botrana* females, probably mainly due to this high concentration of tannins. It may explain why larvae avoid eating the seeds when the size of the berry and their development stage permit.

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References

Awmack CS & Leather SR (2002) Host plant quality and fecundity in herbivorous insects. Annual Review of Entomology 47: 817–844.

Baillod M & Baggiolini M (1993) Les stades repères de la vigne. Revue Suisse de Viticulture Arboriculture Horticulture 25: 7–9.

Balachowsky A & Mesnil L (1935) Insectes nuisibles à la vigne *Polychrosis botrana* Schiff. (Lep. Tortricidae). Les Insectes Nuisibles aux Plantes Cultivées; Leurs mœurs, leur destruction, Vol. 1 (ed. by A Balachowsky & L Mesnil), pp. 677–686. Masson et Cie., Paris, France.

Bovey P (1966) Super-famille des Tortricoidea. Entomologie Appliquée a l'Agriculture, Vol. 2 (ed. by A Balachowsky), pp. 456–893. Masson et Cie., Paris, France.

Bulmer MG (1983) Models for the evolution of protandry in insects. Journal of Theoretical Biology 35: 195–206.

Fagerström T & Wiklund C (1982) Why do males emerge before females? Protandry as a mating strategy in male and female butterflies. Oecologia 52: 164–166.

Geny L, Saucier C, Bracco S, Daviaud F & Glories Y (2003) Composition and cellular localization of tannins in grape seeds during maturation. Journal of Agricultural Food Chemistry 51: 8051–8054.

Harbertson JF, Kennedy JA & Adams DO (2002) Tannins in skins and seeds of Cabernet Sauvignon, Syrah, and Pinot noir berries during ripening. American Journal of Enology and Viticulture 53: 54–59.

Hegarty MP, Hegarty EE & Gentry AH (1991) Secondary compounds in vines with an emphasis on those with defensive functions. The Biology of Vines (ed. by FE Putz & HA Mooney), pp. 287–310. Cambridge University Press, Cambridge, UK.

Kennedy JA, Troup GJ, Pilbrow JR, Hutton DR. Hewitt D et al. (2000) Development of seed polyphenols in berries from *Vitis vinifera* L. cv. Shiraz. Australian Journal of Grape and Wine Research 6: 244–254.

Landolt PJ & Phillips TW (1997) Host plant influences on sex pheromone behaviour of phytophagous insects. Annual Review of Entomology 42: 371–391.

Lederhouse RC, Finke MD & Scriber JM (1982) The contributions of larval growth and pupal duration to protandry in the black swallowtaill butterfly, *Papilio polyxenes*. Oecologia 53: 296–300.

Mackey AP (1978) Growth and bioenergetics of the moth *Cyclophragama leucosticta* Grunberg. Oecologia 32: 367–376.

Maher N, Toulouse ME, Jolivet J & Thiéry D (2000) Oviposition preference of the European grape vine moth, *Lobesia botrana* (Lepidoptera: Tortricidae) for host and non-host plants present in Bordeaux area. Integrated Control in Viticulture IOBC/wprs Bulletin 23(4), 131–134.

Mondy N (1998) Etude du mutualisme entre un champignon phytopathogène *Botrytis cinerea* et un ravageur de la vigne *Lobesia botrana*: recherche de nouvelles méthodes de lutte. Thèse n° 616, University of Bordeaux 2, France.

Mondy N & Corio-Costet MF (2000) The response of the grape berry moth (*Lobesia botrana*) to a dietary phytopathogenic fungus (*Botrytis cinerea*): the significance of fungus sterols. Journal of Insect Physiology 46: 1557–1564.

Moreau J, Benrey B & Thiéry D (2006) Grape variety affects larval and also female performance of the European grapevine moth (*Lobesia botrana*). Bulletin of Entomological Research 96: 205–212.

Peng Z, Hayasaka Y, Iland PG, Sefton M, Hoj P & Waters E (2001) Quantitative analysis of polymeric procyanidins (tannins) from grapes (*Vitis vinifera*) seeds by reverse phase high-performance liquid chromatography. Journal of Agricultural Food Chemistry 49: 26–31.

Quinn GP & Keough MJ (2002) Experimental Design and Data Analysis for Biologists. Cambridge University Press, Cambridge, UK.

Raven CP (1961) Oogenesis: The Storage of Developmental Information. Pergamon Press, Oxford, UK.

Roehrich R & Boller E (1991) Tortricids in vineyards. Tortricid Pests, Their Biology Natural Enemies and Control (ed. by LPS van der Geest & HH Evenhuis), pp. 507–514. Elsevier, Amsterdam, The Netherlands.

SAS Institute (1995) JMP: Statistics and Graphic Guide. SAS Institute, Cary, NC, USA.

Savopoulou-Soultani M, Nikolaou N & Milonas PG (1999) Influence of maturity stage of grape berries on the development of *Lobesia botrana* (Lepidoptera: Tortricidae) larvae. Journal of Economic Entomology 92: 551–556.

Savopoulou-Soultani M, Stavridis DG & Tzanakakis ME (1990) Development and reproduction of *Lobesia botrana* on vine and olive inflorescences. Entomologia Hellenica 8: 29–35.

Savopoulou-Soultani M, Stavridis DG, Vassiliou A, Stafilidis JE & Iraklidis I (1994) Response of *Lobesia botrana* (Lepidoptera: Tortricidae) to levels of sugar and protein in artificial diets. Journal of Economic Entomology 87: 84–90.

Savopoulou-Soultani M & Tzanakakis ME (1988) Development of *Lobesia botrana* (Lepidoptera, Tortricidae) on grapes and apples infected with the fungus *Botrytis cinerea*. Environmental Entomology 17: 1–6.

Slansky F & Rodriguez JG (1987) Nutritional ecology of insects, mites, spiders, and related invertebrates: an overview. Nutritional Ecology of Insects, Mites, Spiders, and Related Invertebrates (ed. by F Slansky & JG Rodriguez), pp. 1–69. Wiley, New York, NY, USA.

Slansky F & Scriber JM (1985) Food consumption and utilization. Comprehensive Insect Physiology, Biochemistry and Pharmacology (ed. by GA Kerkut & LI Gilbert), pp. 87–163. Pergamon Press, Oxford, UK.

Sokal RR & Rohlf FJ (2001) Biometry, 3rd edn. Freeman, New York, NY, USA.

Stavridis DG & Savopoulou-Soultani M (1998) Larval performance and oviposition preference for known and potential hosts by *Lobesia botrana* (Lepidoptera: Tortricidae). European Journal of Entomology 95: 55–63.

Stoeva R (1982) Les hôtes de la teigne bariolée des vignes *Lobesia botrana* Schiff en Bulgarie. Horticultural and Viticultural Science 19: 83–89.

Thiéry D (2005) Vers de la grappe, les connaître pour s'en protéger. Guides pratiques, Vigne et Vin Intl., Bordeaux, France.

Thiéry D & Moreau J (2005) Relative performance of European grapevine moth (*Lobesia botrana*) on grapes and other hosts. Oecologia 143: 548–557.

Thomas AW (1989) Food consumption and utilization by 6th-instar larvae of spruce budworm, *Choristoneura fumiferana*: a comparison on three *Picea* (spruce) species. Entomologia Experimentalis et Applicata 52: 205–214.

Torres-Vila LM & Rodriguez-Molina MC (2002) Egg size variation and its relationship with larval performance in the Lepidoptera: the case of the European grapevine moth *Lobesia botrana*. Oikos 99: 272–283.

Torres-Vila LM, Rodriguez-Molina MC, Roehrich R & Stockel J (1999) Vine phenological stage during larval feeding affects male and female reproductive output of *Lobesia botrana* (Lepidoptera: Tortricidae). Bulletin of Entomological Research 89: 549–556.

Torres-Vila LM, Stockel J & Roehrich R (1995) Le potentiel reproducteur et ses variables biotiques associées chez le mâle de l'eudémis de la vigne *Lobesia botrana*. Entomologia Experimentalis et Applicata 77: 105–119.

Wiklund C & Fagerström T (1977) Why do males emerge before females? Oecologia 31: 153–158.

Wiklund C, Kaitala A, Lindfors V & Abenius J (1993) Polyandry and its effect on female reproduction in the green-veined white butterfly (*Pieris napi* L.). Behavioural Ecology Sociobiology 33: 25–33.

Wiklund C, Nylin S & Forsberg J (1991) Sex-related variation in growth-rate as a result of selection for large size and protandry in a bivoltine butterfly, *Pieris napi*. Oikos 60: 241–250.

Winkler AJ (1962) Development and composition of grapes. General Viticulture (ed. by AJ Winkler), pp. 116–150. University of California Press, CA, USA.