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# Grape cultivar affects larval and female fitness of the European grapevine moth, *Lobesia botrana* (Lepidoptera: Tortricidae)

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**Abstract:** The reproductive performance of phytophagous capital breeder insects is strongly influenced by the food ingested by the juvenile stages and females may adapt their reproductive output in accordance with the diet they ingested as larvae. We studied this for the grapevine pest *Lobesia botrana* (Denis and Schiffermüller), which larvae feed on fruits or berries of different plant species. Larvae were offered one of eight grape varieties as food and their performance and adult reproductive output was measured. The individual performance of *L. botrana* was determined by measuring a suite of life history traits, from egg hatching to adult death. Larval development time, adult mating success, female fecundity, egg size and egg fertility were all significantly influenced by the variety of grape on which the larvae were reared. Interestingly, adult females adapted their reproductive output depending on which variety they had fed and this was correlated with their rate of development. Compared to slow-developing females, females that developed faster laid more eggs, but this was balanced by lower egg fertility. It further presents evidence for diet dependent plasticity in reproductive output in this grape pest. The average reproductive output was highest for females that were reared on cultivars on which larval development was intermediate. Therefore, variation in the performance of *L. botrana* is the result of differential suitability of larval food. This could influence the number of efficient reproductive adults on the next generation, and thus the cultivar on which larvae develops should considered as a factor affecting the amount of European grapevine moth damages.

**Key words:** *Lobesia botrana*, Fitness, population dynamics, grape cultivar, larval food.

## Introduction

The European grapevine moth is one the major pest insect damaging the bunch production in almost all European vineyards. Grape producers are well aware of indirect damage, *L. botrana* larvae favouring the grey mold, but direct damage can also affect the quality of the harvest (Rousseau et al., 2005). Its biology has been rather well described (Bovey, 1966, Gallet 1982, Thiéry, 2005), however the host plant selection process of this polyphagous insect and the consequences of the host selection has received until last decade only little attention. Rather classically, the different grape cultivars receive different level of injury. Several reasons may explain such differences. First, ovipositing females show preferences among among plants and grape cultivars (Maher et al., 2000; Maher et al., 2001, Maher, 2002), these preferences being mainly attributed to the non volatile information present on the berry surface that stimulates oviposition (Maher, 2002; Maher & Thiéry, 2004a; Maher et al., 2006) or to oviposition deterrents produced by competitors or by unsuitable hosts (Thiéry & Gabel, 1993, Gabel & Thiéry, 1996; Callas et al., 2006). The bunch architecture also partly explain differences in the number of larvae installed on the bunches (Fermaud, 1998) and also cultivars show differential suitability for larval installation (Gabel & Roerich, 1995). Several studies stressed out that the grey mould *Botrytis cinerea* added to the larval food could

provide fitness gain to the larvae by reducing the larval development time (Savopoulou-Soultani & Tsanakakis, 1998; Mondy & Coriot-Costet, 2000). Effect of larval food quality could thus be expected to play an important rule in the reproductive performance and thus the population dynamics of that species. This importance of larval food could be reinforced by the fact that several hosts of *L. botrana* may also improve several life history traits in that species (Stavridis & Savopoulou-Soultani, 1998; Thiéry & Moreau, 2005) and that diets enriched with grape seeds play a reverse effect (Moreau et al., 2006).

We postulate thus that the food consumed by the larva is of primary importance for the reproductive success of this species. One may hypothesize that different grape cultivars offers to the larvae of *L. botrana* different nutritional quality and that it has consequences on the overall reproduction of that species. We present here the effect of several grape cultivars on a whole set of life history traits related to *L. botrana* reproduction.

## Material and methods

### *Insects*

The strain of *L. botrana* (INRA Bordeaux) used for this study originated from individuals collected in a French Sauternes vineyard (cultivar white sauvignon). This culture is based on important number of adults caged (several thousands per week) in order to avoid genetic drifts. The stock culture is maintained on a semi-artificial diet at 24 ± 1°C, 60 ± 10% r.h. with a photoperiod of L15:D8 +1 hour of dusk. The first 15-photophase hours were at 1000 lux luminosity and the last one (dusk) at 25 lux. All following tests were performed under these same conditions.

### *Grapes and general procedure*

The grape cultivars were compared using a standardized procedure adapted from (Mondy & Corio Costet, 2000) and similar in principle to that described in Thiéry & Moreau, 2005). This procedure uses an artificial medium in which freeze-dried fine powder of plant material is incorporated. This procedure has at least three main advantages: a) feeding isolated larvae prevents competition and subsequent food deprivations, b) it prevents differences in grape bunch compactness, which has an effect on larval feeding behaviour, and c) it also prevents the incidence of infections by fungi on grapes which affect 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 a medium composed (for 100 Eppendorfs): 150 ml water, 5g agar, 6g cellulose powder, 4g vitamin-free casein, 3.5g glucose, 2g mineral salt, 0.12g cholesterol, 0.12g maize oil, 0.25 benzoic acid, 0.1g nipagine and 12g freeze-dried plant powder. The plant powders were obtained from bunches of *V. vinifera* cv. Bunches of 7 cultivars [Pinot noir (PIN), Chardonnay (NAY), Grenache (GRE), Riesling (RIE), Gewürztraminer (GEW), Merlot (MER) and Chasselas (CHA)] and the wild grape Lambrusque (LAM) were harvested from our "gene collection of grape plants" "Domaine de la Grande Ferrade", INRA-Bordeaux. The bunches were collected at the beginning of the growing season (beginning of May 2003) at phenological stages 23-27 (Eichhorn & Lorenz scale) which correspond to the grape phenology on which the first annual larval generation of *L. botrana* usually feeds. The Eppendorf lids were pierced to allow air circulation. Using a fine brush, newly-hatched larvae (age < 24h) were transferred individually to the diets in each Eppendorf, 100 larvae per diet. Neonate larvae issued from eggs oviposited from thousands of caged females were randomly chosen and attributed to the different diets. Eppendorf tubes were randomized in the Eppendorf racks and racks were moved every three days in the climatic chamber in order to minimize the effect of possible

climatic gradients. All newly emerged adults resulting from the three larval diets were used to evaluate the reproductive output of females. Newly (less than 1 day old) emerged females were individually confined in 0.5 litre transparent cellophane bags as mating and oviposition chambers and provided with water *ad libitum* through a soaked cotton dental wick. One or two days old virgin males originating from the same diet were added to each caged virgin female 1h before dusk, which is just before their sexual activity. Males were randomly assigned to females. Pairs were caged in these bags until the death of both sexes. Females could thus behave and oviposit freely inside the cellophane bag until death.

***Life history traits studied***

Six life history traits (LHT) were monitored in this work:

*LHT 1:* *larval development time* measured as the time between egg hatching and adult emergence. *LHT 2: mating success* determined by the proportion of mated females

*LHT 3:* *female fecundity* measured by isolating couples in translucid bags and by scoring daily the number of eggs laid during the whole female lifespan. *LHT 4: Oviposition period duration* scored as the duration in days from the first to the last egg. *LHT 5: Egg size* measured under binocular using a micrometer lens. Their surface was estimated as an elliptic surface, $S = \pi \times a \times b$ ($mm^2$)), where *a* and *b* are the ellipse semi axes).

*LHT 6:* *egg fertility.* This was scored as the proportion of hatched eggs after 10 days incubated at a temperature of 22°C constant.

## Results

***Larval development time***

Male larvae always developed faster than female ones. The statistical analysis indicates a strong effect of sex but also within each sex of the cultivar on which the larva developed (figure 1).

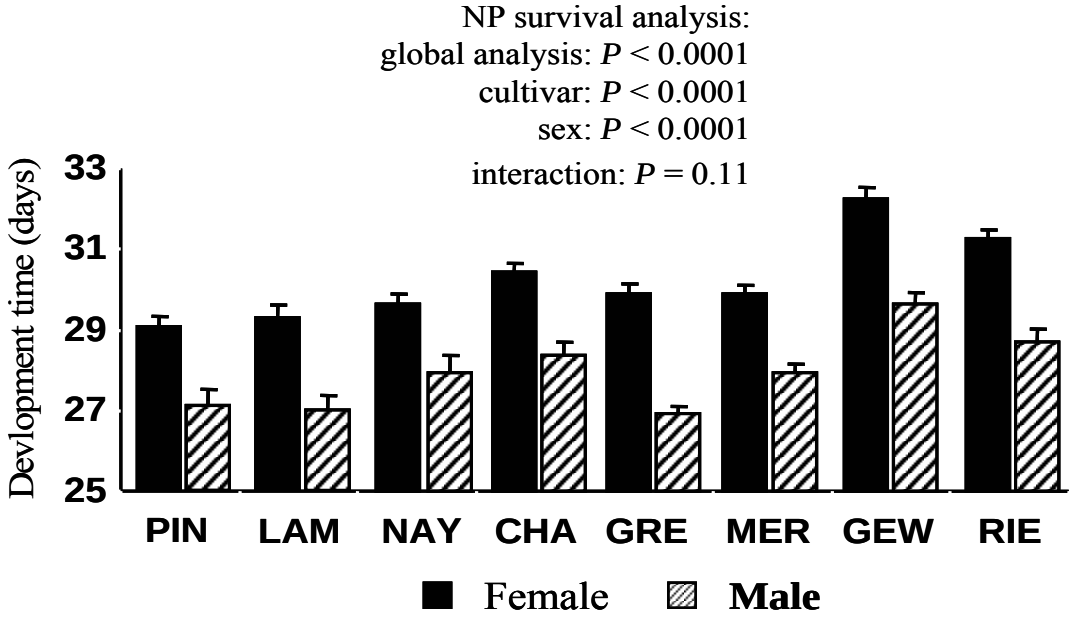


Fig. 1. Development time of male and female *L. botrana* larvae on 8 different grape cultivars. Bars represent mean ± SEM.

This allowed classify the 8 grapes according to the development time of females. Using a hierarchical analysis, 3 groups of development could be identified (Figure2). A first group called ‘slow developer’ includes Gewurtztraminer, Riesling and Chardonnay, a third group called ‘fast developer’ includes pinot noir and Lambrusque, the 3 other grape cultivars being group in an intermediate one.

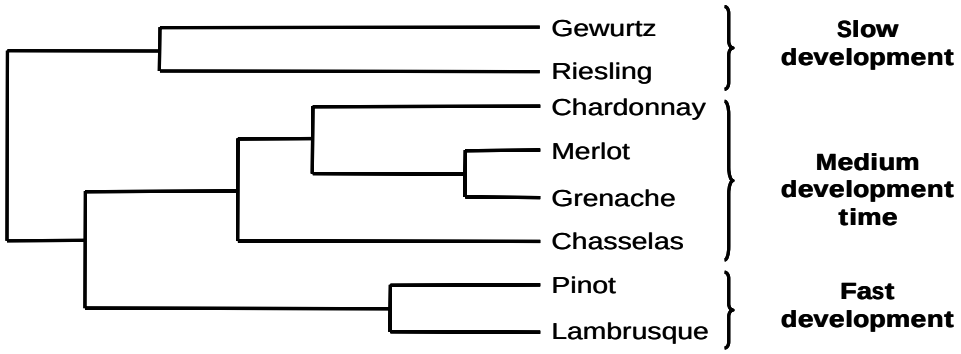


Fig. 2. Classification of the 8 grapes based on the larval development time (cluster analysis based on the ward method).

***Mating success***

The mating success also significantly varied according to the grape cultivar. It ranged from 65.4 % (Riesling) to 97.7 % (Merlot)(Figure 3). The slow developer presented the lowest mating success, and the fast developer were amongst the most successful at mating.

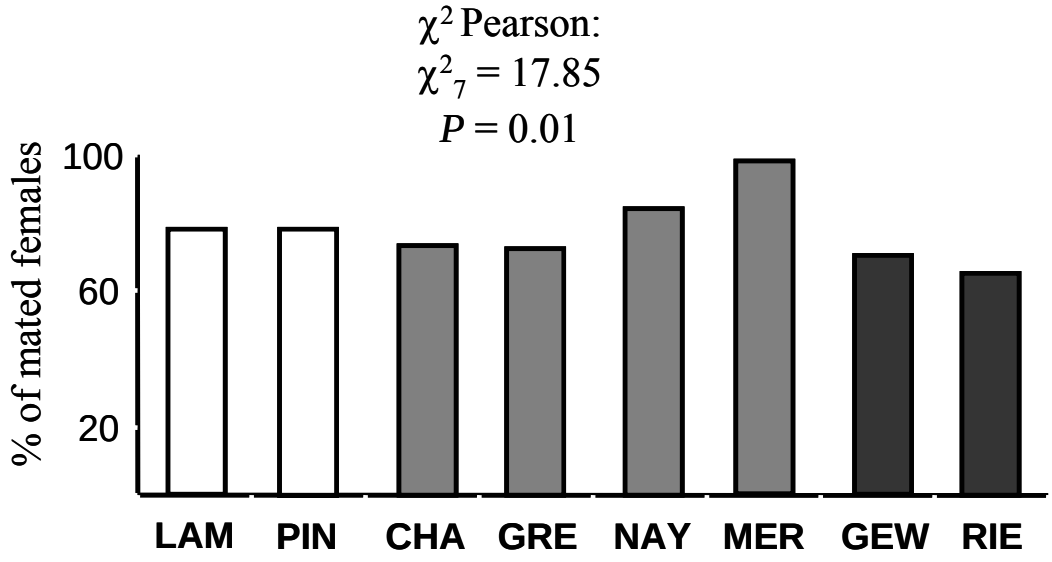


Fig. 3. Mating success of *L. botrana* measured on couples (1 female-1 male) originating both from larvae raised on the different cultivars.

***Female fecundity***

Female fecundity also statistically varied according to the grape cultivar (figure 4). Lambrusque females were the more fecund (mean = 151.2 eggs) and Riesling ones the less (mean = 104.3 eggs). The same tendency was observed as with mating success, the slow developer being the less fecund when the fast ones were amongst the more fecund.

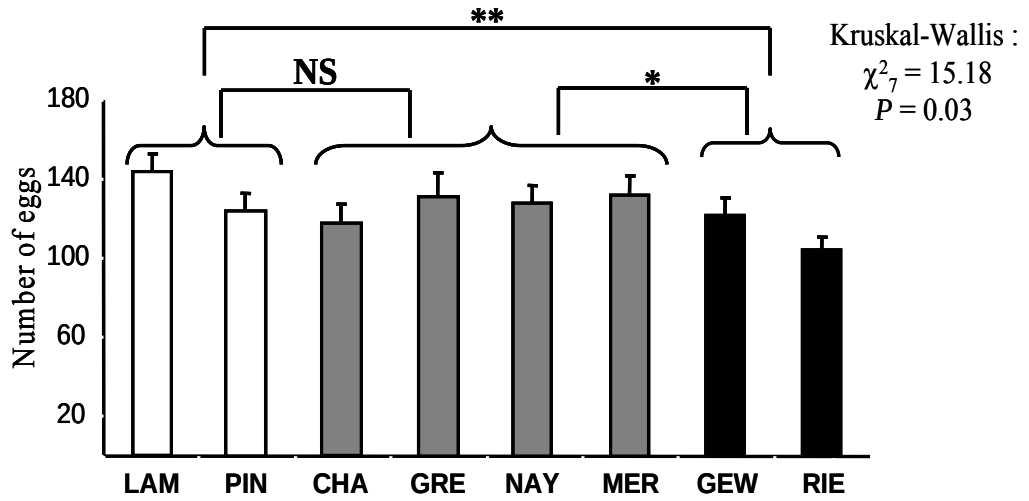


Fig. 4. *L. botrana* female fecundity scored by the number of eggs laid during their lifespan. Values are means ± SEM.

***Oviposition duration***

The grape cultivar also statistically influenced this duration ranging from 4.8 ± .3 days in Riesling to 6 ± .3 days in Pinot noir.

***Egg size***

The result with egg size was reversed to that obtained with the previous LHT. Grape cultivars also induced statistical differences among them, but the bigger eggs were observed with the cultivars that induced the slower development time (Figure 5).

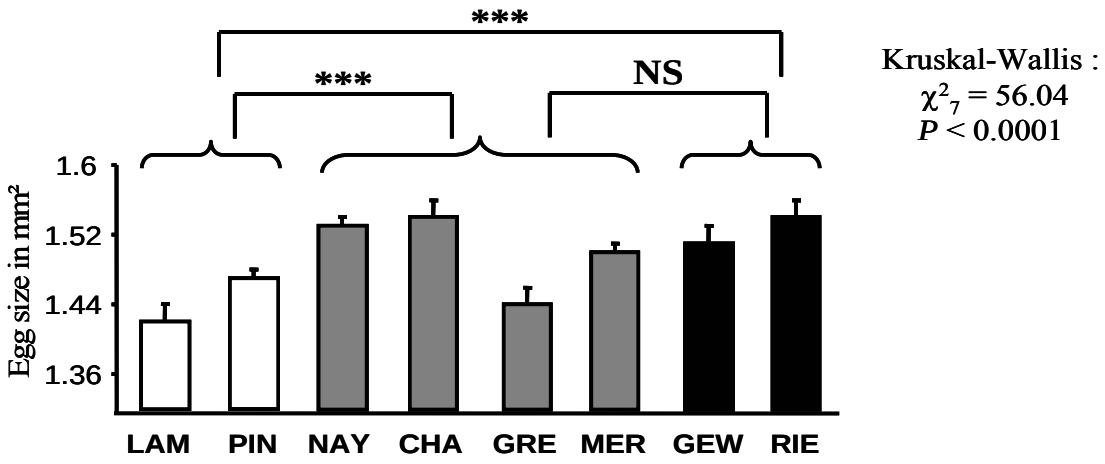


Fig. 5. *L. botrana* egg size according to the cultivar on which larvae were raised. Colours indicate the 3 groups identified figure 2. Values are means ± SEM.

***Egg fertility***

A strong difference among fertilities was observed according to the grape cultivar. This variable is globally well correlated to the egg size which is classical trend in *L. botrana* like in several other moth species. The more fertile eggs were observed in the 'medium' and 'slow' developing groups.

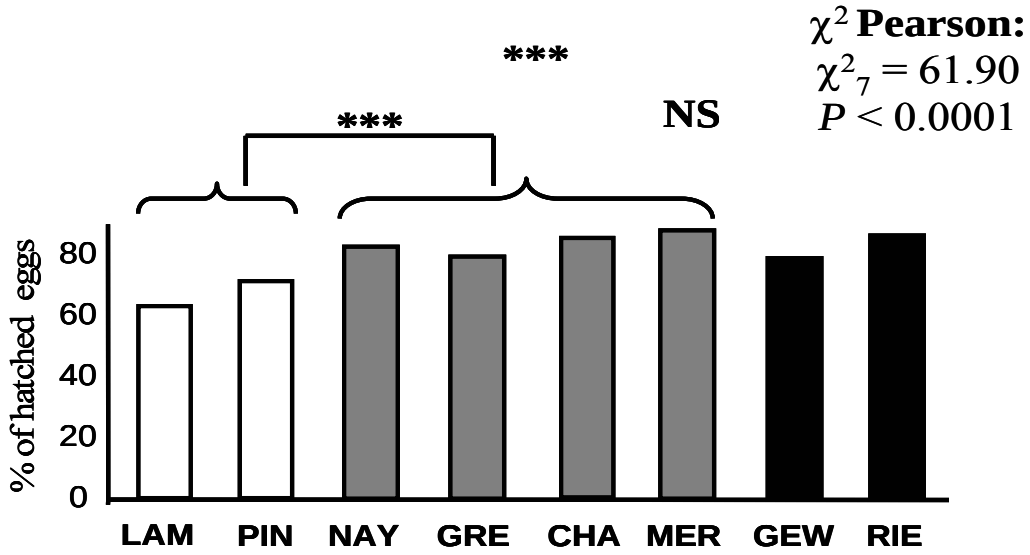


Fig. 6. Proportion of hatched eggs of *L. botrana* according to the cultivar on which the larvae developed. Colours indicate the 3 groups identified figure 2.

## Discussion and conclusion

In the present study, the different grape cultivars generated in *L. botrana* different life history traits related to the reproductive success. This is in agreement with previous studies considering different host plants or grapes infested with the fungus *Botrytis cinerea* (See Thiéry & Moreau, 2005 for a review). This supports that performance of *L. botrana* may vary as a consequence of larval food by influencing the number of efficient reproductive adults and their population dynamics. The differences we have observed help to understand the reason why such a short lived females invest such energy and time at selecting the berry on which it will oviposit (Maher, 2002; Maher & Thiéry, 2004b; Thiéry, 2005). Interestingly several of these life history traits appear in trade off (like fecundity and egg fertility). Therefore, from the list of cultivars we have tested, no 'good' or 'bad' cultivars for *L. botrana* could be qualified. However we hypothesize from this work that several different reproductive strategies could be triggered by the food quality offered here by different grape cultivars: quick development with good mating success and high fecundity but lower fertility and a slow larval development with low mating success and fecundity which are compensated by high egg fertility. Because of these trade offs, the average reproductive output is highest for females reared on cultivars on which larval development was intermediate. This situation seems to occur also in natural populations of *L. botrana* collected from different vineyards (Moreau, Thiéry, Troussard and Benrey unpublished data). At the light of these results, one may expect that population dynamics and thus outbreaks of this insect could vary according the cultivars which are present in certain vineyards areas. We however did not consider here the interactions between cultivar, 'terroir' and climate which can modify the biochemical

expression of the berries and thus its quality as food for the larvae. Also the complete evaluation of the effect of the grape cultivar on the efficiency of the reproductive strategy involves the resistance of the offspring to the parasitoids or predators. This point is currently in study, but these results are not presented here. From the present results, larval food quality provided by the different grape cultivars should be considered for a better understanding of the population dynamics at the next generation. Consequently, this might improve the prognosis of the level of damage due to this species.

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