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Species diversity of larval parasitoids of the European grapevine moth (*Lobesia botrana*, Lepidoptera: Tortricidae): The influence of region and cultivar

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ABSTRACT

The European grapevine moth, *Lobesia botrana* (Denis & Schiffermüller, 1775) (Lepidoptera: Tortricidae) is a major pest of grapevines responsible for great economic losses and frequent insecticide applications. Nevertheless, the use of parasitoids as potential biological control agents has received very little attention. In this study, we present results from a survey on parasitoid species collected from French and Swiss vineyards over a period of 2 years. Over 2000 larvae of *L. botrana* were collected from six grape cultivars out of which a total of 118 parasitoids emerged. Ten species were identified. *Exochus notatus* (Holmgren, 1858) (Hymenoptera: Ichneumonidae) was the most abundant species and has a wide geographical distribution. Another parasitoid species, *Agrypon anxium* (Wesmael, 1849) (Hymenoptera: Ichneumonidae), had never been described on *L. botrana* before and two others remain unidentified *Goniozus* sp. (Förster, 1851) (Hymenoptera: Bethyridae) and *Apanteles* sp. (Hymenoptera: Braconidae). Other species include *Phytomyptera nigrina* (Meigen, 1824) (Diptera: Tachinidae) as well as *Campoplex capitator* (Aubert, 1960), *Diadegma fenestralis* (Holmgren, 1860), *Dicaelotus inflexus* (Thomson, 1891) *Itopectis maculator* (Fabricius, 1775) and *Triclistus meridiator* Aubert, 1984 (all Hymenoptera: Ichneumonidae). We observed substantial regional variation in species richness and also found that abundance and diversity of several parasitoid species varied as a function of geographical location and grape cultivar. The parasitism rate by *E. notatus* was for example affected by grape cultivar suggesting that biological control may be affected by the cultivar of the host plant. These results are discussed in the context of tritrophic interactions and focus especially on the role of grape cultivar on parasitoid richness. We also suggest potential development of native parasitoid species in the implementation of biological control programs against *L. botrana*.

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1. Introduction

In pest management programs aimed to control *Lobesia botrana* (Lepidoptera: Tortricidae), pesticides are used extensively. The most commonly-used compounds are currently insect growth regulators, but techniques such as mating disruption allow a reduction in their use. Because of low control efficacy, development of resistance, and increasing environmental concerns with insecticide use, an alternative to the control of this pest is the implementation of biological control programs using beneficial organisms (Gurr and Wratten, 2000; Eilenberg et al., 2001).

In biological control, one strategy is to favor the use and conservation of native natural enemies already present in the ecosystem rather than introducing alien species. For this type of biological control program to be successful, it is essential to have a good

knowledge of the identity of the natural enemies that are present in the pest's environment, as well as the influence of environmental variation in parasitoid community structure and diversity (Thacker, 2002). In addition, it is crucial to understand the nature and strength of the interactions between the pest, its host plants, and its parasitoids (van Lenteren, 2006).

The ability of a natural enemy to suppress pest populations and hence its efficacy as a biological control agent, may not only depend on its direct interactions with its host, but also on the interactions (direct and indirect) between the natural enemy and the host plant of the pest. Host plant species and varieties differ in their allelochemistry and can affect herbivore and parasitoid performance (Fuentes-Contreras et al., 1996; Turlings and Benrey, 1998; Helms et al., 2004; Thompson et al., 2005; see Ode, 2006 for a review). For example, the survival of *Cotesia melanoscela* (Hymenoptera: Braconidae), a larval parasitoid of the Gypsy moth, is different when larvae are reared from three different plant species (Werren et al., 1992). Similarly, Zvereva and Rank (2003) found that larvae of the

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leaf beetle *Chrysomela lapponica* (Coleoptera: Chrysomelidae) suffer different parasitism rates on different host plants. Some characteristics of the host can also affect the natural enemies directly. For example, while foraging, parasitoids encounter hosts of different sizes and quality and can select the most suitable hosts for the development of their offspring. Many studies have shown that parasitoid species can develop successfully in different sizes of hosts but often prefer a particular size (i.e. Sait et al., 1997; Karamaouna and Copland, 2000; Hanks et al., 2001). Knowledge of these preferences is important to select the appropriate species. Indeed, the efficiency of parasitoids in pest population control depends on their ability to significantly reduce the pest density. If parasitoids attack only the smaller hosts and if these smaller hosts do not actively contribute to the reproductive success, the choice of such a parasitoid species may be inadequate to control the pest. Therefore, knowledge of the effect of host plant and the host selection may contribute to a better understanding of the population dynamics of pests and parasitoid which is requested to develop successful biological programs.

The European grapevine moth *L. botrana* is the major pest of grapes in Europe, North Africa and west Asia (Bovey, 1966; Roehrich and Boller, 1991; Thiéry, 2008) due to its wide geographical distribution and the great damage it may cause to vineyards. In Europe, depending on the region, *L. botrana* completes 2–4 broods a year. The first overwintering adults appear at the beginning of the spring generally when the vine bears few leaves. The duration of a generation varies from 1 to 2 months, depending on the region. Adults of *L. botrana* are mainly active at night (mating starts at nightfall, as well as egg laying which starts 2–3 days after the first mating). The first generation of eggs is laid on the flower bud, sometimes on the bracts, the vine-shoots or the leaves, those of later generations on the grapes. The first instars present a “strolling stage” of only few hours, and then bore into the flower bud (called glomerulae). The caterpillar starts its development at the time of flowering, the first generation finishing its development on small green berries. The second generation emerges at the end of June–July and the third generation occurs between mid-August and the end of September. Larvae are polyphagous and can develop on almost all cultivars of grapes and other plant species (Stoeva, 1982; Roditakis, 1988; Savopoulou-Soultani et al., 1990; Thiéry, 2008). Larval food quality, including different host plants and grape cultivars, has been shown to result in variable reproductive performance of adult moths (Thiéry and Moreau, 2005; Moreau et al., 2006a,b, 2007). Each generation may cause serious damage to bunches either by quantitative but also qualitative losses especially by facilitating the infection of pathogenic fungi like the grey mold disease *Botrytis cinerea* Persoon: Fries (Leotiales: Sclerotiniaceae) (Roehrich and Boller, 1991) or black mold *Aspergillus* spp. (Thiéry, 2008). Therefore, important economic losses due to that pest or different fungi related to larval feeding on berries are regularly attributed to the grapevine moths (Thiéry, 2008).

In the absence of effective biological control measures, vineyards are mainly treated with pesticides. In French vineyards, between two and five pesticide applications are generally recommended against *L. botrana*. In recent years, great emphasis has been placed on mating disruption for control of grape berry moth infestations with some success (Charmillot et al., 1995). Biological control is one possible future strategy against this pest but has received little attention. Several studies have reported the occurrence of different species of parasitoids and predators on *L. botrana* in vineyards (Marchesini and Dalla Monta, 1994; Coscolla, 1997; Schirra and Louis, 1998; Perez Moreno et al., 2000; Thiéry et al., 2001; Xuéreb and Thiéry, 2006, and see also Thiéry, 2008 for a review). To our knowledge only one experimental study has examined the interactions between parasitoids, their hosts and their host plants in vineyards (Xuéreb and Thiéry, 2006). In this study, conducted on the five

main grape cultivars in the Bordeaux region, parasitism rates were not affected by the grape cultivar on which the host developed but was positively correlated with the host density.

From the information presented above, it is clear that in order to implement a successful biological control program against *L. botrana*, more information is needed on the potential biological control agents. The goal of the present study was to conduct a survey of parasitoid diversity and abundance in several vineyards in Switzerland and France. We were specifically interested in answering the following questions: (1) which larval parasitoid species are naturally occurring in vineyards attacking the first generation larvae of *L. botrana* and what is their respective abundance? This information is basically nonexistent for vineyards in Switzerland (Genini, 2000), (2) are the same parasitoid species occurring in different geographical regions? (3) Is their distribution and abundance associated with the type of cultivar? For the most abundant parasitoid species, we also studied variation within each geographic region.

2. Materials and methods

Vineyards were selected for several reasons: (i) large plots containing multiple grape varieties offering a choice both for the host and the parasitoids in each vineyard and (ii) practical reasons for easier samplings. Larval collections were done when population levels of *L. botrana* were over 30 larvae per 100 bunches in each cultivar. Within each vineyard, the different grape varieties received identical viticultural conditions but variation in soil conditions were not considered in this work. With this procedure, we were able to measure the direct effect of cultivar within a vineyard while avoiding potential confounding effects due to environmental variation (temperature, light exposure, humidity) that exists between vineyards.

In France, two vineyards were sampled in June 2003 (corresponding to the end of the first moth generation) at Tavel in the south and at Colmar in the east and one vineyard in 2004 in the south of France (at Roquemaure). In Switzerland, two vineyards were sampled in June of 2003 (at Yvorne in the canton Vaud and at St Pierre-de-Clages in the canton Valais), and in June of 2004 (at Nyon in the canton Vaud and at Sion in the canton Valais) (see Fig. 1). In each site, if the population density was sufficient, we sampled in different grape cultivars (Fig. 1).

We sampled larvae from the first generation of *L. botrana* in young flower buds. Larvae appear inside silk nests called glomerulae. Larvae of *L. botrana* almost never move from one bunch to another; each collected larva was therefore considered as having accomplished its whole development on this bunch (Torres-Vila et al., 1997). Collections were not destructive, only the silk nest with the larvae inside was removed from the bunch. At each site, all nests were examined and only late instars (L4 and L5 instars) and pupae were gathered. Larvae were kept in small boxes (15 × 10 × 12 cm) with berries and fed *ad libitum* until the end of their development.

In the lab, larvae were checked daily until pupation. Pupae were then extracted from the berries, weighed to the nearest 0.01 mg and placed individually in glass tubes (70 mm × 9 mm diameter) closed with cotton plugs, labeled and stored in the room at 23 °C and under natural photoperiod. Pupae collected in the field were put directly in the glass tubes. Pupae were checked daily for adult emergence. All emerged parasitoids were preserved in 70% ethanol. Parasitism was measured *a posteriori* by checking emergence and was calculated per cultivar as the percentage of parasitism [(number of parasitoids/(number of *L. botrana* + number of parasitoids emerged)] × 100). Identification of parasitoids was based on adult morphological criteria. In each cultivar, around 10% of pupae did

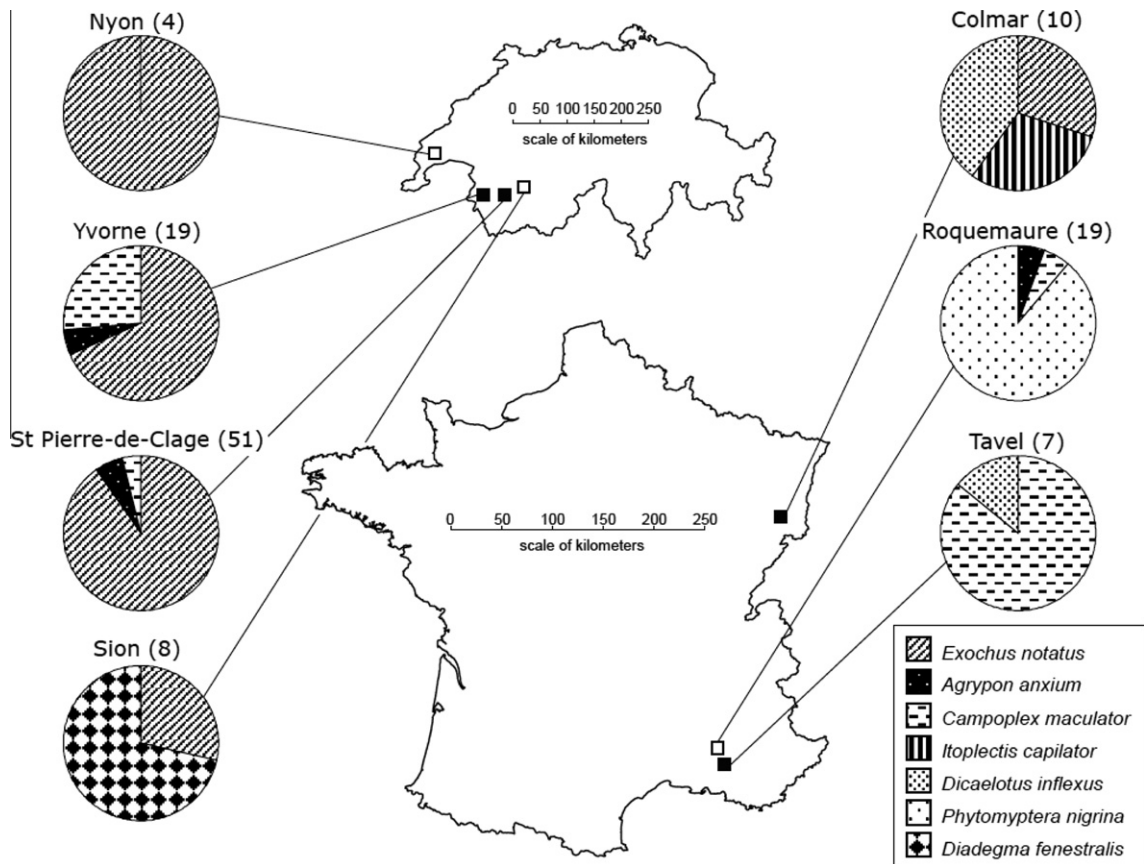


Fig. 1. Map showing the localities in France and Switzerland sampled in 2003 (black squares) and 2004 (white squares) and the relative abundance of parasitoid species attacking *L. botrana* in each site. The rare parasitoid species are not represented in the figure. The numbers in bracket represent the number of parasitoid found.

not emerge. No dissection was attempted to determine non-emerged pupae to establish if there is a *L. botrana* of parasitoid inside.

2.1. Statistical analyses

All statistical tests were performed using JMP software (Version 3.2.2, SAS Institute Inc.) and two-tailed tests of significance were used throughout. Data from the 2 years of surveying were analysed independently of each other. In order to test if the total parasitism rates varied according to the geographical region and cultivars for each year, a logistic regression was conducted where the cultivar factor was nested within geographical region (Zar, 1999; Samuels and Witmer, 2003). Pearson χ^2 tests were used to compare the total parasitism rate between different cultivars within the same vineyard. Finally, we wanted to see if there was an effect of pupae mass on the emergence of either *L. botrana* or a parasitoid species. As pupal mass depends on the cultivars on which larvae fed (Moreau et al., 2006a,b, 2007), we conducted this analysis only within the same cultivar and only on the more abundant parasitoid species. We used an ANOVA followed by Tukey's least significant difference procedure to test the difference between groups (male moth, female moth or parasitoid) as the assumptions for parametric analyses were met (Shapiro–Wilk's test for normality and Levene's test for homogeneity of variances) (Sokal and Rohlf, 1995).

3. Results

3.1. Parasitism species

During the 2 years of surveying, 2471 *L. botrana* larvae were collected from which a total of 118 parasitoids emerged (Table 1). Ten

species were identified, and they differed in both abundance and distribution (Table 1 and Fig. 1). The ichneumonid, *Exochus notatus* was the most abundant and represented 57% of the parasitoids collected in both years. This species was found everywhere except in the south of France. The tachinid *Phytomyptera nigrina* was the second most abundant parasitoid species but was found only in the south of France. The third most abundant parasitoid species was *Campoplex capitator* (Ichneumonidae) which was found everywhere except in the east of France. The other parasitoid species, found at a lower abundance (less than 10 individuals) were: the ichneumonids *Diadegma fenestrale*, *Dicaelotus inflexus*, *Itoplectis maculator*, *Agrypon anxium* and *Triclistus meridiator*, the braconid *Apanteles* sp. and the bethylid *Goniozus* sp. (Table 1).

3.2. Parasitism rate

The mean parasitism rate was significantly higher in 2003 (10%) than in 2004 (2%) (Pearson $\chi^2 = 88.41$, $P < 0.001$) (Fig. 2). During the first year (2003), the parasitism rate varied among locations and also among the cultivars (Nested Nominal logistic, Whole Model: $\chi^2_3 = 43.97$, $P < 0.0001$, effect of location: L-R $\chi^2_3 = 27.84$, $P < 0.0001$, effect of cultivars nested in location: L-R $\chi^2_4 = 13.84$, $P = 0.008$) (Fig. 2a). The parasitism rate was very low in the east of France and higher in the south of France and in the canton of Valais in Switzerland. Within the two geographical sites in Switzerland, larvae collected on Pinot cultivars had a higher probability of being parasitized than larvae collected on Chasselas or Gammay (Table 2). However, no difference was observed in the east of France between the two cultivars (Table 2). In addition, for the most abundant parasitoid species (*E. notatus*), we tested the effect of cultivar within the same vineyard (St Pierre-de-Clages) on the

Table 1 Parasitoid species identified during the 2-year survey in different geographic regions in France and Switzerland and their abundance according to the cultivar sampled (VD: Vaud; VS: Valais; SF: South France; EF: East France).

Year	Locality	Cultivar	<i>Exochus notatus</i> (Ichneumonidae)	<i>Phytomyptera nigrina</i> (Tachinidae)	<i>Campoplex capitator</i> (Ichneumonidae)	<i>Agrypon anxium</i> (Ichneumonidae)	<i>Diadegma fenestralis</i> (Ichneumonidae)	<i>Dicaelotus inflexus</i> (Ichneumonidae)	<i>Itoplectis maculator</i> (Ichneumonidae)	<i>Apanteles</i> sp. (Braconidae)	<i>Triclistus meridator</i> (Ichneumonidae)	<i>Goniozus</i> sp. (Bethyliidae)
2003	Yvorne (VD)	Pinot noir	6		2	1						
	Yvorne (VD)	Chasselas	7		3							
	St Pierre-de-Clages (VS)	Pinot noir	18									
	St Pierre-de-Clages (VS)	Gamay	8			1						
	St Pierre-de-Clages (VS)	Chasselas	20		2	2						
	St Pierre-de-Clages (VS)	Chasselas	20		6		1	1				
2004	Tavel (SF)	Grenache	2				1		2		1	
	Colmar (EF)	Gewurtz Riesling	2				1	1	1			
	Colmar (EF)	Grenache	2				3					
	Roquemaure (SF)	Grenache		16								
	Roquemaure (SF)	Syrah			1	1				1		
	Sion (VS)	Pinot noir										1
Total	Sion (VS)	Chasselas	2				5					
	Nyon (VD)	Chasselas								1		
	Nyon (VD)	Chardonnay	3			5	5	5	3	2	1	1
Total			66	16	14	5	5	5	3	2	1	1

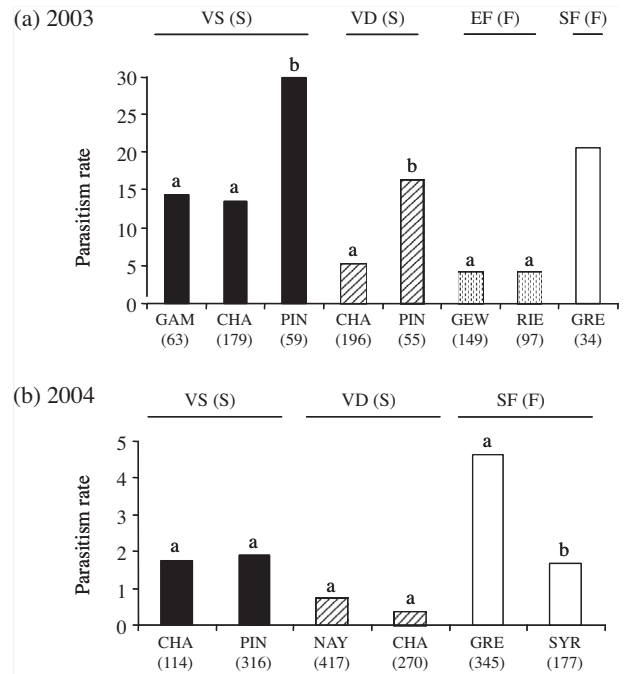


Fig. 2. Results on parasitism rate found (a) 2003 and (b) 2004 in different localities (VS: Valais; VD: Vaud; EF: East France; SF: South France; S: Switzerland; F: France) and on different grape cultivars (GAM: Gammay; CHA: Chasselas; PIN: Pinot; GEW: Gewürztraminer; RIE: Riesling; GRE: Grenache; NAY: Chardonnay; SYR: Syrah). Bar colors are arranged according to the locality of sampling. Columns sharing the same letter are not significantly different ($P > 0.05$) after a Pearson χ^2 test in the parasitism rate between cultivars within the same vineyard. The numbers in bracket represent the number of pupae of *L. botrana* sampled with emergence of either a moth or a parasitoid.

Table 2

Results from the comparison of parasitism rate between two cultivars within the same vineyard. Line in bold indicates those statistically different.

Year	Locality	Comparison of cultivar	Pearson χ^2	P
2003	St Pierre-de-Clages (VS)	Gammay vs Chasselas	0.03	0.86
		Gammay vs Pinot	7.4	0.006
		Chasselas vs Pinot	3.8	0.05
Yvorne (VD) Colmar (FE)	Chasselas vs Pinot	7.78	0.005	
	Gewurtztraminer vs Riesling	0.001	0.97	
2004	Roquemaure (FS)	Syrah vs Grenache	3.10	0.04
	Sion (VS)	Chasselas vs Pinot	0.01	0.92
	Nyon (VD)	Chardonnay vs Chasselas	0.34	0.55

parasitism rate. This species parasitized more *L. botrana* larvae in Chasselas and Pinot noir than those found in Gammay (Pearson $\chi^2 = 12.65$, $P = 0.002$) (Table 1).

For the second year (2004), the parasitism rate varied again among sites but no effect of cultivar was detected (Nested Nominal logistic, Whole Model: $\chi^2_5 = 19.12$, $P = 0.001$, effect of location: L-R $\chi^2_2 = 8.64$, $P = 0.01$, effect of cultivars nested in site: L-R $\chi^2_3 = 3.65$, $P = 0.30$) (Fig. 2b). The parasitism rate was higher in the south of France than in Switzerland. Despite the non significant effect of cultivars shown in the full model, we detected a cultivar effect when we restricted our analyses to the south of France (Table 2). Indeed, larvae fed on Grenache cultivar had a higher probability of being parasitized than larvae fed on Syrah.

3.3. Mass of *L. botrana* pupae and parasitism by different species

We restricted our mass analyses to the three most abundant parasitoid species namely, *E. notatus*, *P. nigrina* and *C. capitator* and to cultivars on which we collected enough larvae to perform statistical analyses (Fig. 3). For all cultivars, females of *L. botrana* emerged from larger pupae than males did (ANOVA, $F_{2,342} = 204.36$, $P < 0.0001$ for Grenache 2004; ANOVA, $F_{2,30} = 6.21$, $P = 0.006$ for Grenache 2003, ANOVA, $F_{2,56} = 12.10$, $P < 0.0001$ for Pinot noir 2003, ANOVA, $F_{2,172} = 60.46$, $P < 0.0001$ for Chasselas 2003). In the south of France, a post hoc test showed that *P. nigrina* and *C. capitator* parasitoids were recovered mostly from smaller hosts (Fig. 3). However, no effect of pupal mass was revealed for *E. notatus*.

4. Discussion

In this study, we found that (1) parasitism rate strongly varied between years and geographic regions, (2) within a vineyard, parasitism rate also varied according to the cultivar, (3) our results indicated the presence of one main species, *E. notatus*, across different geographic regions except in the south of France, (4) a considerable regional variation was found in species composition, and (5) there is a host-size dependent parasitism for some parasitoid species.

Ten species of larval parasitoids of *L. botrana* were recorded but only two dominant species appear to be good candidates for natural biological control of *L. botrana*. The most promising candidate appears to be *C. capitator* because this solitary larval endoparasitoid has a wide geographical distribution and it has been regularly observed in most of the European vineyards (Italy, Spain, Switzerland, France) (Marchesini and Dalla Monta, 1994; Coscolla, 1997; Genini, 2000; Thiéry et al., 2001; Thiéry, 2008). In our study, this species was the third in term of density but in most of other surveys of *L. botrana*, it appeared as the most abundant parasitoid species (Marchesini and Dalla Monta, 1994; Thiéry et al., 2001; Thiéry and Xuéreb, 2003). For these reasons it is considered the main larval parasitoid of *L. botrana* (Marchesini and Dalla Monta, 1994; Coscolla, 1997; Genini, 2000; Thiéry et al., 2001; Thiéry and Xuéreb, 2003; Xuéreb and Thiéry, 2006; Thiéry, 2008) but our data seems to contradict this trend. The capability of *C. capitator* to parasitize diapausing pupae of *L. botrana* indicates that they can parasitize the third generation of *L. botrana* larvae, which occurs in late autumn (Thiéry, unpublished data). Because of its natural efficiency, density and wide geographical distribution, it may provide an important natural control of *L. botrana* population. A release of this parasitoid species early in the season may reduce reproduction of further generations of the moth.

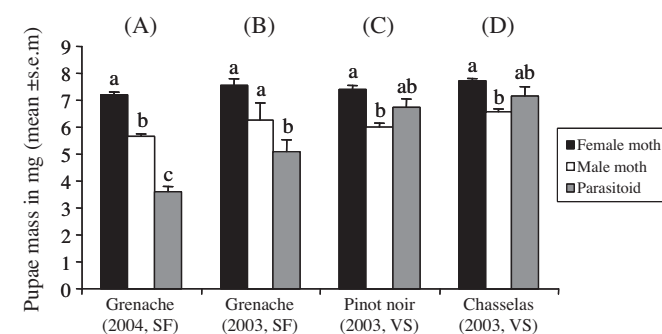


Fig. 3. Mass of *L. botrana* pupae from which emerged either (i) a female moth, (ii) a male moth or (iii) a parasitoid species for a given cultivar. (A) *Phytomytera nigrina* in Grenache (B) *Campoplex capitator* in Grenache, (C) *Exochus notatus* in Pinot noir, (D) *Exochus notatus* in Chasselas. Columns sharing the same letter are not significantly different ($P > 0.05$) after a parametric PLSD test.

One second candidate is the parasitoid *E. notatus*. It was the most abundant species in our study, representing 57% of the parasitoids collected in both years. It has also a wide geographical distribution (except in the south of France). This observation was to some extent surprising since most previous samplings conducted on *L. botrana* often reported *C. capitator* as the major species as noted above. However, *E. notatus* has been previously recorded as a parasitoid of *L. botrana* in Germany (Schwangart, 1918), Bulgaria (Zapryanov, 1985) and Russia (Telenga, 1934). Another species, *E. tibialis* (Holmgren, 1858), a close relative of *E. notatus*, was also recorded on *L. botrana* by Schwangart (1918) and has been collected, although in low numbers, on this host during the last decades in Italy (Pinna et al., 1989; Marchesini and Dalla Monta, 1994). Because of its significant parasitism rates and wide geographical distribution, *E. notatus* may, as *C. capitator*, be an efficient biological control agent against *L. botrana*.

The tachinid *P. nigrina*, was the second most abundant parasitoid species, but it was collected only in the south of France and appears to be restricted to the Mediterranean area (French, Italian, Greek and Turkish vineyards) as confirmed by previous findings (Luciano et al., 1988; Kara and Tschorsnig, 2003; Bagnoli and Lucchi, 2006; Thiéry et al., 2006; Martinez et al., 2007; Thiéry, 2008). Due to its geographical restriction, *P. nigrina* does not appear as a universal candidate in European vineyards except in the southern area. Several other parasitoid species were found (*D. fenestrata*, *D. inflexus* and *I. maculata*), but do not appear to be good candidates for a biological control against *L. botrana* due to either the weak abundance observed (less than 10 individuals) or the restricted geographical distributions. In addition, we report four other marginal species among which one, *Triclistus meridiator*, was also obtained from *L. botrana* and *Argyrotaenia ljugiana* (Thunberg, 1797) near Colmar in 2002 (Kuntzmann and Villemant, unpublished data) and one other *A. anxium* is (as *D. fenestrata*) a well known antagonist of the grape leafroller *Sparganothis pilleriana* (Denis & Schiffermüller, 1775) (Voukassovitch, 1924; Vidal, 1997; Thiéry et al., 2001). Two other parasitoids that belong to the genera *Apanteles* and *Goniozus* need, for identification at species level, the examination of a larger number of specimens.

The most interesting result found in this study is the effect of cultivars on overall parasitism rate inside two vineyards in 2003. In both Valais and Vaud area, larvae collected on Pinot cultivars had a higher probability of being parasitized than those collected on other cultivars. In addition, the most abundant parasitoid species (*E. notatus*) parasitized more *L. botrana* larvae on Chasselas and Pinot noir than on Gammay. Levels of parasitism have frequently been observed to differ between plant species or cultivars (i.e. Helms et al., 2004; Kahuthia-Gathu et al., 2008). This is the first report of an effect of cultivars on the susceptibility of larval parasitism in *L. botrana*, although it was previously shown on egg parasitism during laboratory experiment (Moreau et al., 2009). This cultivar effect was surprising since previous studies have reported that parasitism rate of *C. capitator* did not depend on the grape cultivar offered to the host larvae (Xuéreb and Thiéry, 2006). However, in the study of Xuéreb and Thiéry (2006), there was a strong host density effect: the number of parasitoids that emerged was correlated to the total number of hosting larvae. Here, we only sampled in vineyards that had a rather high larval density of *L. botrana*. These two studies taken together could explain the overall parasitism rates found for *L. botrana*. Firstly, the density of *L. botrana* larvae was most likely the main factor attracting parasitoid species. Parasitoids are known to often use chemical cues for the location of their hosts (Vet and Dicke, 1992; Turlings and Benrey, 1998 for a review) and the presence of larvae at high densities may increase the intensity of chemical cues and as a result, increase the natural parasitism rates (Lessells, 1985; Costamagna et al., 2004; Xuéreb and Thiéry, 2006). Recently, Chucho et al.

(2006) have shown that the larval parasitoid *Dibrachys cavus* (Walker, 1835) (Hymenoptera: Pteromalidae) is attracted to frass produced by *L. botrana* and *E. ambiguella* larvae. Besides host density, the type of cultivar may also influence parasitism rates. Several studies have shown the influence of host plants on parasitism rates in the field (Fuentes-Contreras et al., 1996; Thompson et al., 2005; Helms et al., 2004; Ode, 2006 for a review and references therein). Plant species or cultivars may influence parasitism rates directly or indirectly (Turlings and Benrey, 1998; Cortesero et al., 2000). Hosts developing on different cultivars may differ in their development time such that prolonged development may extend the window of vulnerability of these hosts and result in higher parasitism rates (Benrey and Denno, 1997). In our system this does not appear to be the case. Moreau et al. (2006a,b) found that the length of larval development for larvae fed on cultivar suffering either low (Chasselas) or high (Pinot) parasitism rate was identical. Larvae developing on different cultivars may affect the herbivore ability to mount an effective immune response against natural enemies explaining in turn the variation in parasitism rate. Such effect of plant quality on the herbivorous immune response has already demonstrated (Ojala et al., 2005; Kapari et al., 2006; Klemola et al., 2007; Karimzadeh and Wright, 2008). For example, Klemola et al. (2007) found that individuals of the autumnal moth (*Epirrita autumnata*, Lepidoptera Geometridae) reared on naturally low-quality food showed a higher ability to mount an effective immune response as compared to those reared on higher quality food. This hypothesis requires experimental confirmation in *L. botrana* by assessing the strength of the larvae immune system fed on different cultivars.

Another interesting result is the effect of pupal mass on parasitoid identity. We found that *P. nigrina* and *C. capitator* emerged from smaller pupae found in Grenache. Two hypotheses may explain this result and some experiments are clearly needed to distinguish between them. First, parasitoids usually encounter hosts of different sizes and it has already been shown that different parasitoid species exploit different size-hosts more effectively than others (Sait et al., 1997; Chau and Mackauer, 2001; Hanks et al., 2001). Therefore, we could hypothesize that these two parasitoid species preferentially attack the smaller hosts. For example, it has already been shown that small larvae may be less capable of physically defending themselves from parasitization than larger larvae or have a less efficiency immune system (Van Alphen and Drijver, 1982). Alternatively, a possibility remains that there is no active choice of a particular host size made by parasitoid species. Indeed, the fact that parasitoids emerged from smaller hosts could be due to the slowly growth of host due to the energy diverted by parasitoid for its own development. Distinguishing between these two alternative hypotheses is crucial since this result can have a profound effect for the success of biological control. For example, *C. capitator* is considered by some authors to be an optimal candidate for controlling *L. botrana* populations (Xuéreb and Thiéry, 2006; Thiéry, 2008). If this species preferentially attacks the smaller hosts, the timing for field release of these species should be set to when the younger larvae are most abundant, whereas if the second hypotheses are true, the timing of releases are not important. Therefore, knowledge of the host preference would lead to a better understanding of the population dynamics of the host and parasitoid and could improve the success of biological program.

In conclusion, the results from this survey of parasitoid species of *L. botrana* in vineyards from different regions indicate that the right species to use in a biological control program will vary with the geographic region. In addition, its efficiency may be affected by the cultivars on which *L. botrana* larvae feed. Thus, a successful biological control program against *L. botrana* should not only consider the pest and the potential parasitoid, but also the interactions between the parasitoid, their hosts and host plant in different habitats.

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