

# Seasonal Fluctuation, Percent Damage and Larval Natural Enemy of Grape Berry Moth (*Lobesia botrana*) in Tehran Province (Shahriar Region) and Ghazvin Province (Takestan Region), Iran

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## ABSTRACT

Grape berry moth, *Lobesia botrana* (Lepidoptera: Tortricidae) is one of the most important pests in vineyards around the world. Since the extent of its damage and the role of natural enemies have not yet been deeply studied in Iran, we decided to assess the situation of this pest in the Takestan region in Ghazvin Province. In this research, we showed that grape berry moth has three generations in this area and that damage caused in the third generation is more extensive than in either the first or second generation. In addition, we identified one new parasitoid larva (*Enytus apostata*). To produce healthy grapes, we should apply alternative pest control methods like mating disruption, mass trapping and biological control (e.g. inoculation of *Trichogramma cacoeciae*).

**Keywords:** *Enytus apostata*, parasitoid, *Trichogramma cacoeciae*

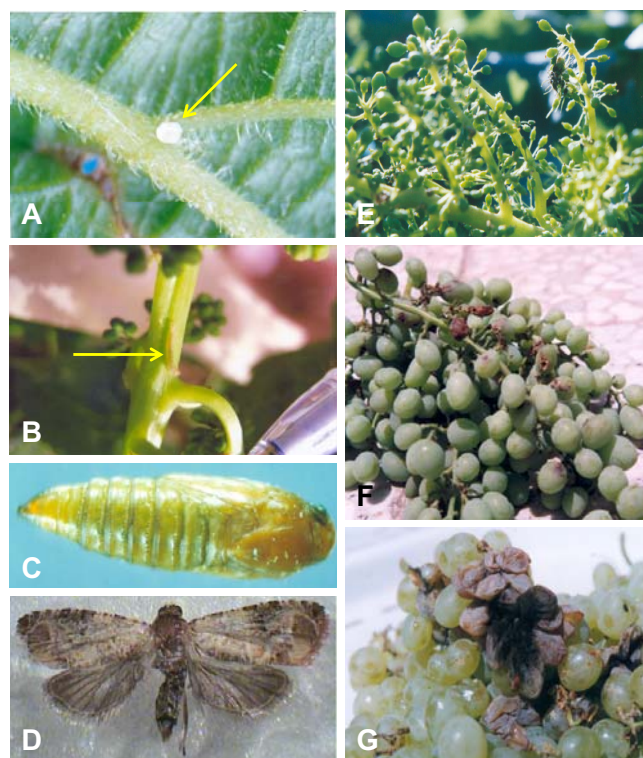
## INTRODUCTION

Grape berry moth (GBM), *Lobesia botrana*, is one of the most important pests of vineyards in Iran and worldwide (e.g. Oliva *et al.* 1999; Varandas *et al.* 2004). GBM reduces the quality and quantity of grape fruit, raisin and vinegar market value drastically.

A description of this pest and its life-cycle is as follows. Eggs: The eggs are flat and elliptical (Crop Protection Compendium 2000). Eggs: The eggs are flat and elliptical. Egg dimensions are approximately 0.8 mm × 0.6 mm. Eggs are laid singly (Fig. 1A) or in groups of 2-3. Larvae: There are 5 instars. Larval size ranges from 1 to 15 mm. Larval color ranges from light green to brown, depending on the nutrition (Fig. 1B). Pupae: Pupa size ranges from 4 mm to 9 mm. Pupa coloration is initially light brown and later changes to dark brown (Fig. 1C). Adult: Adult body length ranges from 6 mm to 8 mm and the wingspan ranges from 10 mm to 13 mm. Body coloration is cream with black marks. The legs exhibit alternating white and brown bands. The forewings exhibit a mosaic pattern of black, brown, white, blue and red. The hind wings are brownish-gray (Fig. 1D).

The alternate hosts for *L. botrana* include: cherry, Chinese gooseberry, carnation, persimmon, blackthorn, pomegranate, red currant, plum, black currant and olive (Venette *et al.* 2004). GBM damages grapes by feeding on flower buds and fruits, and feeding results in external and internal damage and subsequent rotting due to fungal pathogens. One of the most serious infections is gray rot, which is caused by *Botrytis cinerea* (Fermaud 1990). This damage results in lower crop yield, lower grape quality and bad bouquet flavor in the case of wine grapes.

This pest possesses 3 to 4 generations annually, depending on regional climatic conditions. The first generation



**Fig. 1** Grape Berry Moth (*Lobesia botrana*) life-cycle. (A) Egg under the leaf surface; (B) Larva; (C) Pupa; (D) Adult moth; (E) Damage during the first generation; (F) Damage during the second generation; (G) Damage during the third generation.

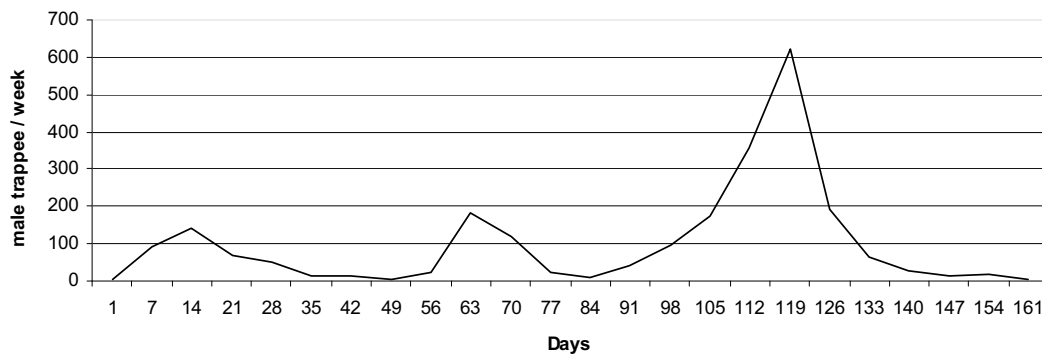


Fig. 2 Seasonal fluctuation of GBM in the Shahriar region, Iran in 2004.

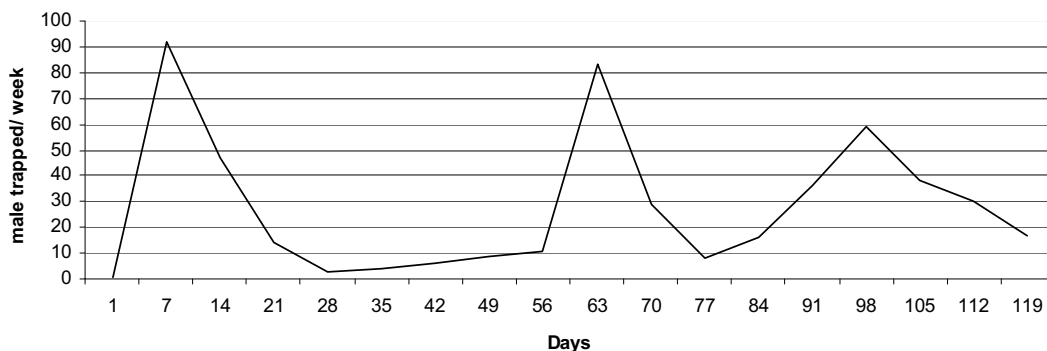


Fig. 3 Seasonal fluctuation of GBM in the Takestan region, Iran in 2005.

feeds on grape inflorescences and the second generation feeds on sour grape berries. The third and fourth generations feed on sweet grape berries (Figs. 1E-G).

An understanding of the feeding behavior of this pest allows for the modeling of the emergence time of moths, thus allowing for more focused treatment and control (Moravie *et al.* 2006).

The amount of damage this pest inflicts is significantly increased when the vine is also infected by *Botrytis cinerea* (grey mold) (Mondy *et al.* 1998) and the third generation is of greatest economic importance (Fermaud and Giboulot 1992). The spores of this fungus disseminate by *L. botrana* larvae into the berries (Fermaud and le Mean 1992).

The primary objective of this study was to estimate the percent damage of GBM in vineyards in Iran and to identify natural enemies of the larva.

## METHODOLOGY

To determine the number of GBM per generation, Delta pheromone traps (Tehran Plant Protection Organization) at one meter above the ground in white seedless orchard grape (*Vitis vinifera* cv. 'Soltani') were installed. Moths trapped were identified according to the descriptions of Nasr *et al.* (1995) and Fowler and Larkin (2002) and counted twice a week.

The percent damage of GBM in vineyards was calculated in five different orchards (each about one hectare), almost one kilometer away from each other. In these orchards no control measures were taken against GBM. Thirty trees of each orchard were randomly selected (one tree in each row), then all clusters of those trees were counted.

With the completion of the *L. botrana* life-cycle, the number of damaged grape clusters were counted (9 clusters from 35 trees).

Sampling was non-destructive.

In another, separate experiment, the percent damage/larva in grape clusters was determined. This test examined only one orchard in the Takestan region in an area in which no pesticides had been applied. Five infested clusters from each of thirty trees from the north, south, east, west and center of each tree were selected and at the end of each generation the number of damaged inflorescences, sour berries and sweet berries of selected clusters were counted. Experiments were completely randomized and the means were compared by a factorial test.

To find larval natural enemies, damaged clusters with larvae were collected, and were reared in insectariums to provide pupae. Specifically, larvae were placed in glass dishes and every 48 h, a

fresh grape cluster was placed in the glass because fungal diseases tend to attack the berries in the insectarium. The conditions inside the insectarium were:  $25 \pm 5^\circ\text{C}$ ,  $50 \pm 5\%$  RH and  $16 \pm 8$  h photoperiod.

Larvae were then separated from each other. Some larvae in the first or second instar were parasitized by *Enytus apostata* (Hymenoptera: Ichneumonidae). Finally adult wasps emerged from pupae instead of adult GBMs. These parasites were placed in 70% alcohol in a test tube and were sent to Dr. Hannes Baur, Department of Invertebrates, Natural History Museum (Switzerland) and then to Prof. Klaus Horstmann (Zoological Institute, Wurzburg University, Germany) for final identification.

## RESULTS AND DISCUSSION

Our study showed that GBM had three generations per year in the Shahriar region in 2004 (Fig. 2). The first catches of GBM were in late April and the peak of the adult population for the first, second and third generations were in early May, late June and late July, respectively. In the Takestan region in 2005, the first catches were in mid-April, and the peak of the adult population for the first, second and third generations were in late April, late June and mid-August, respectively (Fig. 3; Table 1).

In the Takestan region in 2005, the lowest percent damage by GBM was shown by the first generation (2.9%) and the highest values in the third generation (50.8%) (Table 1).

The parasitoid wasps that appeared in our insectariums belonged to the Ichneumonidae family, and following identification and classification by Dr. Hannes Baur and Prof. Klaus Horstman, were considered to be *Enytus apostata* (Gravenhorst 1829), subfamily Campopleginae, which was reported in Iran for the first time (Fig. 4A, 4B).

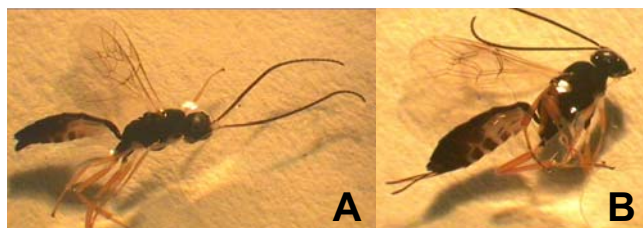


Fig. 4 Male (A) and female (B) parasitoid.

**Table 1** The percent damage of GBM in five different orchards on three generations.

Orchard №	Generation	Percent damage*
1	I	2.883 h
1	II	8.669 f
1	III	42.272 b
2	I	3.001 h
2	II	6.234 g
2	III	23.692 d
3	I	7.242 fg
3	II	12.489 e
3	III	42.879 b
4	I	7.254 fg
4	II	14.375 e
4	III	50.780 a
5	I	4.181 gh
5	II	6.452 g
5	III	37.849 c

\* Data in the column are significantly different when followed by a different letter according to the LSD test ( $p < 0.05$ ).

**Table 2** Percent damage of GBM in clusters per larva in Ghazvin province, Takestan region in 2005.

Generation	Percent damage*
I	7.23 b
II	4.00 c
III	11.33 a

\* Data in the column are significantly different when followed by different letters according to the LSD test ( $p < 0.05$ ).

Our study showed that the percent damage in cluster on third generation is more than the first and second generation, because on third generation, *Botrytis* fungi is attack to the berries and the percent damage of GBM on the second generation in clusters is less than in either the first or third generations (Table 2). In contrast, the amount of pest activity in the second generation was more than in the first generation although the percent damage in clusters was less. It is believed that the amount of acidity of sour grape berries negatively affects the feeding of *L. botrana* (Torres-Vila *et al.* 1995).

In vineyards in which pesticides were not applied, the percentage damage by the second generation is about two times higher than that caused by the first generation and the percent damage by the third generation was about four times higher than that caused by the second generation and eight times more than the first generation (Table 1). In addition, the population activity of GBM is more than their natural enemies. There are a few reports about other natural enemies of GBM, the most important of which are *Trichogramma evanescens* (Hym., Trichogrammatidae) (Nasr *et al.* 1995) and *T. cacoeciae* (Barnay *et al.* 1999).

The most conventional method to control this pest is to apply commercially available insecticides such as Larvin 375 at a rate of 1.0 L/ha, Bulldock 25 EC at 0.3 L/ha, Enduro 258 EC at 0.3 L/h, Reldan 40 EC at 1.25 L/ha, Polytin 200 EC at 0.1 L/ha, Karate Zeon at 0.25 L/ha, Decis 25 EC at 0.3 L/ha, Total 60 EC at 1.5 L/ha, or Calypso 480 EC at a rate of 100 mL/ha (Torres-Vila *et al.* 1997). In organic farming the spray program is based on a product whose

active ingredient is *Bacillus thuringiensis* (e.g. Agree<sup>®</sup>, Bactospeine<sup>®</sup>, Dipel<sup>®</sup>, BMP 123<sup>®</sup>, wettable powder). In the first generation of GBM, sprays are generally not recommended but for second and third generation usually two treatments are performed (Torres-Vila *et al.* 1997).

To produce healthy grapes, we should apply alternative pest control methods like mating disruption, mass trapping and biological control.

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