

Research Article

Effects of three *Quercus* species on feeding performance of the green oak leaf roller, *Tortrix viridana* L. (Lepidoptera: Tortricidae)

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Abstract: Feeding performance of the green oak leaf roller, *Tortrix viridana* (Linnaeus) was determined on three host plants including *Quercus libani* Oliv, *Quercus infectoria* Oliv, and *Quercus branti* Lindl, under laboratory conditions. Mean total leaf weight consumed by larvae was the highest on *Q. branti* (0.31 g) and the lowest on *Q. libani* (0.14 g). The feeding index was the highest on *Q. libani* while the other two host plants were not significantly different. Significant relationships were found between amount of consumed leaf tissues and resulting pupal weight, on all host plants. Larval and pupal mortality were not significantly different on host plants. Findings indicated that the feeding index could be a key factor to assess performance of *T. viridana* on *Quercus* species.

Keywords: *Tortrix viridana*, host plants, feeding index, consumption rate

Introduction

The green oak leaf roller *Tortrix viridana* L. is one of the most important defoliators on oaks in many European, Asia Minor and Northern African countries (Kalapanida and Glavendekic, 2002; Schroder and Degen, 2008). This tortricid is a monophagous pest of oak stands in western Iran (Fazeli and Abaei, 1990). Females of *T. viridana* lay majority of their eggs on branches close to next year's buds of the host oaks. The insect overwinter as eggs on the branches, and hatch next spring (Hunter, 1990; Du Merle, 1999; Invashov *et al.*, 2002). As this pest is

monophagous on *Quercus*, it may severely damage deciduous oaks, as well as evergreen species (Du Merle, 1999). In oak forests of western Iran, the larvae cause heavy defoliation on three main oak species including *Quercus libani*, *Q. infectoria* and *Q. branti* (Ghobari, 2007). As a consequence of defoliation, the wood increment decelerates with economic loss for the tree stock in commercial forestry (Schroeder and Degen, 2008).

Development of efficient strategies for controlling green oak leaf roller will require knowledge of its biological relationships with various host plants. Among these, an important component will be an understanding of host suitability. Quantitative analysis of consumption and utilization of host plants by insect herbivores is a commonly used tool in studies of plant-insect

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interactions (Scriber and Slansky, 1985). Previous studies on *T. viridana* have demonstrated the role of host plant phenology in the developmental time of this insect pest (Ivashov *et al.*, 2002; Ghirardo *et al.*, 2012). Study of the effect of food on the biology of insects is critical to understanding host plant suitability for herbivore species. As far as we know there are no reports on food utilization, growth, development and survival of *T. viridana* on *Q. libani*, *Q. branti* and *Q. infectoria*. Therefore, this research was undertaken to determine green oak leaf roller larval consumption rates on three common host plants. Also the effects of consumption rate on pupal weights in addition, effects of host plants on developmental times of larvae and pupae of green oak leaf roller were determined.

Materials and Methods

Host plants

The three host plant species examined were *Q. branti*, *Q. infectorian*, and *Q. libani*. The leaves were collected from local wild plants from the oak forest located in Marivan, Kurdistan province in west Iran (35°31'37"N 46°10'35"E). The selected species were based on their importance as the main oak species in forests of north Zagros and being the main host plants of *T. viridana*.

Insect rearing

In early April 2014, twigs from each of the three host plant with overwintering eggs were collected from natural habitat several days before emergence of leaves and were transferred to the Natural Resource Institute at Kurdistan University in separate plastic bags. Twigs were kept in plastic containers of water under outdoor conditions, until the emergence of leaves and were checked for hatched larvae every day. The newly hatched larvae were set upon buds that had just opened using a small brush. They were individually placed on each bud of their respective host plants. The smaller twigs were

placed petri dishes (12 cm diameter) containing 1.5 ml distilled water to avoid desiccation. The whole plastic containers were transferred to a growth chamber at 26 ± 2 °C, $65 \pm 5\%$ RH and 16:8 h (L: D). Normally, the 1st instar larvae hide within the buds until they molt to 2nd instar. At this stage, the larvae come out and look for fresh buds. The buds were replaced by new ones every four days. The larvae were fed with oak leaves collected from the three host plants every second day. At the 4th instar stage, the larvae were transferred into plastic boxes (14 cm × 10 cm × 5 cm) where they also pupated.

Feeding tests

Larval feeding on the three different host plants was carried out by placing one newly hatched larva on an undamaged leaf taken from each of three host plants. The test was undertaken in 8 replications for each of the three host plants. The leaf was placed inside a plastic petri dish (9 cm diameter), while its petiole was covered in moistened cotton to prevent desiccation and kept in a growth chamber at 26 ± 2 °C and photoperiod of 16: 8 h (L: D). After 24 h, excreta were removed and weighed. Petri dishes were changed and fresh leaves were weighed and introduced to larvae. The experiment was conducted throughout larval feeding process and continued until emergence of pupae. To determine weight loss caused by desiccation, the weighed leaves were kept in a petri dish in the similar condition for the same period without larvae and were reweighed after 24 h. Daily food consumption per larva was estimated by subtracting weight of remaining leaf tissue from weight of the provided leaves. The weight of feces produced by the larvae was recorded daily for each oak species. To find the dry weights of the leaf, feces, and larvae, extra specimens (20 specimens for each) were weighed, oven-dried (48 h at 50 °C), and then re-weighed to establish a percentage of their dry weight. All pupae were weighed and their sex was determined. A feeding index was calculated by dividing mean pupal weight by mean weight of leaf tissue consumed for each host plant. Time for

development from egg to adult was recorded for each individual. Insect development was monitored every 6 h, and every 15 min during molting (Kalapanida and Glavendekic, 2002).

Statistical Analysis

The data was subjected to one-way analysis of variance (ANOVA) using SPSS (version 19.0). Means were compared using Tukey's HSD post-hoc test. To examine the correlation between the amount of consumed diet and pupal weight, simple linear regression analysis was used.

Results

Larval consumption rates:

Total consumption rate of *T. viridana* larva on *Q. libani* was significantly lower than two other host plants ($P < 0.01$). The highest amount of food consumption was recorded on *Q. branti* (0.31 g) and the lowest on *Q. libani* (0.14 g) (Fig.1) ($F = 90.8$, $df = 2$, 47 , $P < 0.01$). However, no significant difference was observed on larva fed on *Q. branti* and *Q. infectoria*.

Effects of consumption on pupal weights and larval mortality

Regression analyses showed significant effect of consumed food on pupal weight (Fig. 2). The parallel line regression analyses also revealed significant effect of consumed food on pupal weight with coefficient of determination (r^2) explained ranging from 48% on *Q. libani* to 84% on *Q. infectoria* (Fig. 2). The highest pupal weight gained per unit of diet consumed (113.6 mg/g) was recorded on *Q. libani* (Fig. 2), whereas the lowest on *Q. infectoria* (68.7 mg/g) (Fig. 2).

The mean pupal weight did not differ significantly among the three host plants. However, the larvae fed on *Q. branti* indicated the highest amount of pupal weight and the lowest amount on *Q. libani* ($F = 2.24$, $df = 2$, 17 , $P = 0.141$) (Fig. 3).

The feeding index was highest in *Q. libani* and the other two host plants were not significantly different ($F = 14.175$, $df = 2$, 17 , $P < 0.01$) (Fig. 3).

The larval and pupal mortality reared on three different host plants was not significantly different (Larvae: $F = 0.231$, $df = 2$, 29 , $P = 0.795$; Pupae: $F = 0.088$, $df = 2$, 29 , $P = 0.916$) (Table 1).

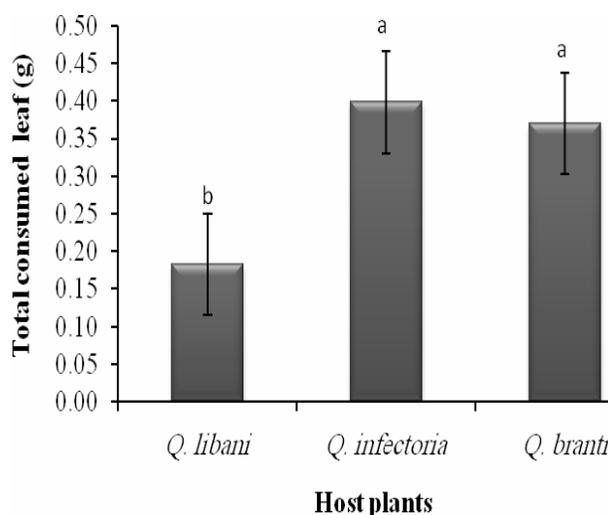


Figure 1 Consumption of three *Quercus* species by larvae of *Tortrix viridana* (bars indicate SE). ($F = 22.667$, $df = 2$, 47 , $p < 0.01$). Means with the same letter are not significantly different (Tukey HSD, $p < 0.05$).

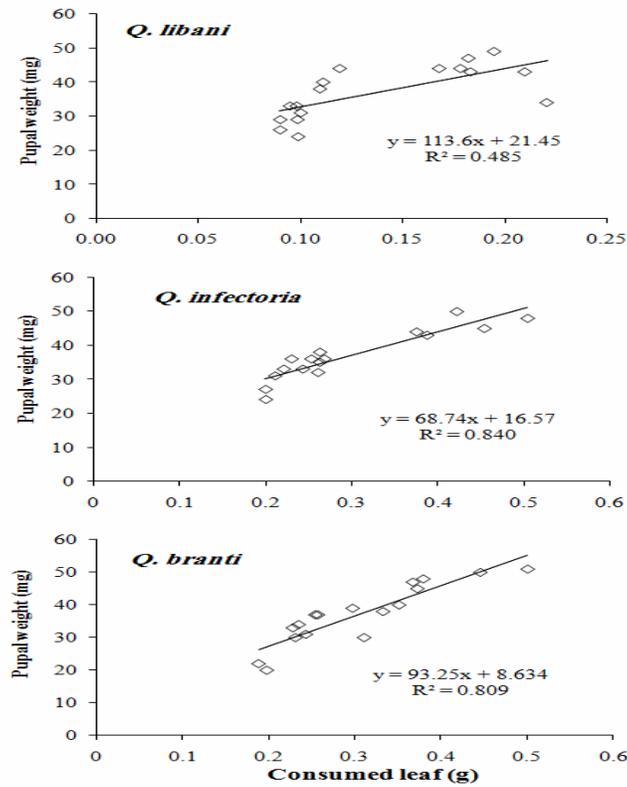


Figure 2 Relationship between pupal weight and consumed food of three *Quercus* species by *Tortrix viridana* larvae.

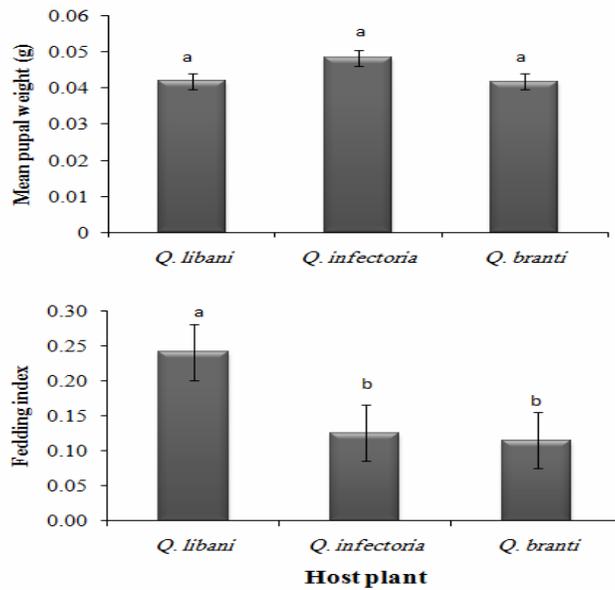


Figure 3 Mean pupal weight and feeding index (pupal weight / consumed food) of *Tortrix viridana* on three *Quercus* species. Error bars indicate SE, Means with the same letter are not significantly different (Tukey HSD, $P < 0.05$).

Table 1 Mortality of *Tortrix viridana* in larval and pupal stages on different *Quercus* species.

Host plant	Mortality (%) ¹		
	Larval stage	Pupal stage	Total
<i>Q. branti</i>	52.4	18.3	70.7
<i>Q. infectoria</i>	51.9	18.8	70.7
<i>Q. libani</i>	51.4	19.1	70.5

¹ The larval and pupal mortality reared on host plants was not significantly different.

Effects of host plants on larval and pupal development

Larval and pupal developmental times are significantly dependant on the host plants during larval feeding (Table 2). Duration of larval stage was longest on *Q. libani* (12.5d) ($F = 36.25$; $df = 2, 29$; $P < 0.01$), but there was no significant difference between larvae fed on the other two host plants. Total developmental time was also the longest on *Q. branti* (19.1 d), intermediate on *Q. infectoria* and the shortest on *Q. libani* (16.8 d) ($F = 9.28$, $df = 2, 29$, $P < 0.01$).

Table 2 Effects of different *Quercus* species on developmental time of *Tortrix viridana*.

Host plants	Development time (day) ¹	
	Larval	Total
<i>Q. branti</i>	12.5 ± 0.22a	19.0 ± 0.31a
<i>Q. infectoria</i>	10.1 ± 0.23b	18.0 ± 0.33b
<i>Q. libani</i>	10.1 ± 0.23b	16.8 ± 0.46c

¹ Means (± SE) within a column followed by the same letters are not significantly different (Tukey HSD, $P < 0.05$).

Discussion

Green oak leaf roller *T. viridana* larval feeding efficiency was evaluated on leaves of three host plants in western Iran. The data clearly showed that *T. viridana* performed differently in feeding efficiency, larval and pupal development, survival and pupal weight when *Q. libani*, *Q. infectoria* and *Q. branti* were

offered as food plants for their larvae. These results are supported by a few studies, for example, Ivashov *et al.* (2002) examined average weights of *T. viridana* pupae on early, normal and late flushing oak trees through four years. Insects feeding on late flushing oaks, gained the highest pupal weights. Average weights of pupae on late flushing oaks were different through four years. It was between 30.4 – 41.7 mg for male insects and 42.9 – 53.1 mg for female insects. In this study, mean pupal weights ranged from 41.8 mg on *Q. branti* to 45.8 mg on *Q. infectoria*. However, pupal weights were not found to be significantly different between host plants. The results of the current study showed that weight of leaf consumed was significantly different among host plants. The larvae fed on *Q. libani* consumed the least amount of food; the lowest pupal weight was expected for this host plant. However, pupal weights of larvae fed on *Q. libani* and *Q. branti* were practically the same. Ghirardo *et al.* (2012) obtained the same results. They investigated larval consumption rates and pupal weight of *T. viridana* on two genotypes of *Q. robur* (tolerated and susceptible oaks). Larvae that developed on T-oaks required significantly more leaf material than that they developed on S-oaks. However, pupal weight did not differ between the two food sources, being on average 33 mg. These results are supported by our previous report where the amount of consumed food of *T. viridana* fourth instar larvae was the least on *Q. libani* (Yazdanfar *et al.*, 2015). However, the greatest amount of larval weight gained was observed in this host plant. These results were probably due to either higher nutritional quality of *Q. libani* for *T. viridana* than that for the other host plants or lower amount of secondary substances. Greenberg *et al.* (2001) examined feeding performance of *Spodoptera exigua* (Hübner) on different host plants. They showed that although total larval consumption was lowest on pigweed, pupal weight was the highest in this plant. In their view, pigweed was of higher nutritional quality for *S. exigua* than the other tested plants.

The shorter larval development time and the lower mortality rate the more suitable the host plant (Awmack and Leather, 2002). In the current study, larval developmental time was longest on *Q. branti* (12.5 d), followed by *Q. libani* and *Q. infectoria* (10.1 d). In addition, total developmental time (first instar to pupae) was longest on *Q. branti* (19.1 d) and was shortest on *Q. libani*. Ghirardo *et al.* (2012) reached almost the same results. They reported that the average timespan of development from the 1st instar to the pupae stage was 17.1 ± 1.9 d for the larvae developed on susceptible *Q. robur* and 16.8 ± 1.6 d for the larvae developed on tolerant *Q. robur*. Kalapanida and Glavendekic (2002) found longer larval developmental times of *T. viridana* through six years. Larval development times ranged from 20.6 to 25.3 d and pupal stage ranged from 9 to 9.7 d. The observed difference in duration of larval and pupal stages of *T. viridana* in the different studies is probably attributed to the climatic conditions and especially to various host plants.

Larval and pupal survival of *T. viridana* was not affected by feeding on different host plants. Kalapanida and Glavendekic (2002) found that survival of *T. viridana* in larval and pupal stage was higher than what we recorded in the current study. They investigated the survival of larvae and pupae through six years and found that the survival of larvae varied from 93.33 to 80.83% and the pupae from 90.65 to 82.83%. However, Ghirardo *et al.* (2012) reported the survival rate of larval development on susceptible and resistant *Q. robur* was 25 and 31%, respectively. This finding is similar to our study.

The effects of different host plants on *T. viridana* were assessed with more focus on feeding index (Fig. 3). Because the amount of consumed food and final pupal weight are simultaneously considered in calculating feeding index, as both parameters are very important in analyzing effects of host plants on insects.

Therefore, the present results concluded that *Q. libani* provides the best food quality for *T. viridana*. These results could probably be due to higher nutritional quality or lack of defensive

compounds in this plant. Levels of nitrogen, total protein, total carbohydrate and water content were highest on *Q. libani* compared to two other host plants, which made it as the more preferred host plant for *T. viridana* larvae (Yazdanfar *et al.*, 2015).

In conclusion, this study demonstrated that *T. viridana* larval performance is significantly better when fed on *Q. libani* than the two other host plants.

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تأثیر سه گونه‌ی بلوط روی تغذیه و بیولوژی پروانه برگ‌خوار بلوط، *Tortrix viridana* L. (Lepidoptera: Tortricidae)

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چکیده: شاخص غذایی، نرخ مصرف و بیولوژی پروانه‌ی برگ‌خوار بلوط، *Tortrix viridana* (Linnaeus) روی سه میزبان بلوط شامل *Quercus libani* Oliv، *Quercus infectoria* Oliv و *Quercus branti* Lindl در شرایط آزمایشگاهی بررسی شد. بالاترین مقدار میانگین برگ خورده شده توسط لاروهای برگ‌خوار بلوط روی میزبان *Q. branti* (۰/۳۱ گرم) و پایین‌ترین مقدار آن روی میزبان *Q. libani* (۰/۱۴ گرم) بود. شاخص غذایی (یعنی وزن شفیره تقسیم بر وزن کل برگ خورده شده) روی میزبان *Q. libani* بالاترین مقدار بود درحالی‌که دو میزبان دیگر از این نظر اختلاف معنی‌داری با هم نشان ندادند. در هر سه میزبان ارتباط معنی‌داری میان مقدار برگ خورده شده و وزن شفیره‌ها، مشاهده شد. زمان نمو لاروها و شفیره‌های برگ‌خوار بلوط روی سه میزبان، متفاوت بود. به‌طوری‌که، کوتاه‌ترین زمان نمو لاروها روی برگ‌های *Q. libani* (۲۶/۹ روز) و طولانی‌ترین زمان روی برگ‌های *Q. branti* (۳۱/۶ روز) بود، زمان نمو روی *Q. infectoria* (۲۸/۱ روز) بود. زنده‌مانی لاروها و شفیره‌ها روی سه میزبان مختلف اختلاف معنی‌داری با هم نشان نداد. شاخص غذایی به‌عنوان یک آماری به‌منظور ارزیابی عملکرد لاروها روی سه میزبان مختلف، استفاده شد. بر این اساس، *T. viridana* بهترین عملکرد را روی گونه *Q. libani* و ضعیف‌ترین عملکرد را روی گونه *Q. branti* از خود نشان داد.

واژگان کلیدی: *Tortrix viridana*، میزبان، شاخص غذایی، نرخ مصرف