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Natural enemies of *Orgyia trigotephras* (Boisduval 1829) (Lepidoptera, Erebidae, Lymantriinae) in Tunisia

Sonia Hammami[a,b,*], Olfa Ezzine[a,b], Samir Dhahri[a], Mohamed Lahbib Ben Jamâa[a]

Abstract: Phytophagous insects may select plants or plant parts not only based upon nutritional content, but also on the intensity of predation and parasitism. This observation leads to the enemy free space hypothesis, which suggests that herbivores select host plants for which the mortality from natural enemies is minimized, by preferring host plants on which the herbivores would be less vulnerable to their natural enemies. In Tunisia, *Orgyia trigotephras* is a polyphagous moth, bivoltine with a spring (SG) and an autumnal (AG) generation. This work was carried out in two sites: Jebel Abderrahmane (Ftahiz, Delhiza and Guitoun) and Bizerte (Sejnane). The aim is to compare the importance of predation by moth larvae (*Coccidiphila rungsella*) and parasitism by egg parasitoids (*Aprostocetus sp.*) on *O. trigotephras* on two host species, *Quercus coccifera* and *Pistacia lentiscus*.
Keywords: *Orgyia trigotephras, Aprostocetus* sp.*, Coccidiphila rungsella, Quercus coccifera, Pistacia lentiscus,* Tunisia

Tunus'ta *Orgyia trigotephras* (Boisduval 1829) (Lepidoptera, Erebidae, Lymantriinae)'ın doğal düşmanları

Özet: Bitki ile beslenen böcekler, sadece besin içeriğine göre değil aynı zamanda avcılık ve parazitlik yoğunluğuna göre bitkileri veya bitkilerin belirli kısımlarını seçebilir. Bu gözlem sonucunda, düşmansız alan hipotezi ortaya atılmıştır. Bu hipoteze göre, otoburlar doğal düşmanlarına karşı daha az savunmasız olabilecekleri konukçu bitkileri tercih ederek doğal düşmanlardan kaynaklanan ölümlerin en az düzeyde olacağı bitkileri seçmektedir. Tunus'ta *Orgyia trigotephras,* ilkbahar nesli (İN) ve sonbahar nesli (SN) ile bivoltin ve polifag bir böcek türüdür. Bu çalışma, iki bölgede gerçekleştirilmiştir: Jebel Abderrahmane (Ftahiz, Delhiza ve Guitoun) ve Bizerte (Sejnane). Bu çalışmanın amacı, Tunus'ta her neslin *Quercus coccifera* ve *Pistacia lentiscus* üzerinde böceğin larvalarının (*Coccidiphila rungsella*) avcılık düzeyi ve *O. trigotephras* yumurta parzitoidlerinin (*Aprostocetus sp.*) parazitlik düzeyinin önemini karşılaştırmaktır.
Anahtar kelimeler: *Orgyia trigotephras, Aprostocetus* sp.*, Coccidiphila rungsella, Quercus coccifera, Pistacia lentiscus,* Tunus

1. Introduction

Phytophagous insects may select plants or plant parts not only based upon nutritional content, but also on the intensity of predation and parasitism. This observation lead to the enemy free space hypothesis, which suggests that herbivores will select host plants for which the mortality from natural enemies will be minimized, by preferring host plants on which the herbivores would be less vulnerable to their natural enemies. *O. trigotephras* is a polyphagous insect. Its abundance varies strongly across regions, from rare and endangered (Dionisio, 2002) to a common defoliator or even to pest status (Villemant and Fraval, 1993; Chakali et al., 2002; Ezzine et al., 2010). *O. trigotephras* is bivoltine, with a spring generation, from April to June and an autumn generation, from October to December (Ezzine et al., 2014). An outbreak of *O. trigotephras* occurred in Tunisia in 2005 and lead to the complete defoliation of more than 500 ha of Jebel Abderrahmane forest (2000 ha) (Ezzine et al., 2010). Egg masses including about 140 eggs are laid in a loose silk net spun between two or three leaves of the host tree (Ezzine et al., 2010).

During the outbreaks, Lepidoptera defoliators cause considerable damage leading to complete defoliation of oak, from which it results in adverse effects on their production and regeneration. Dynamics of these lepidopteran pests is limited by the effect of parasitoids. Each development phase of the life cycle of the insect is preyed by a number of parasitoids or predators (Villemant, 2003), which may be either Hymenoptera or Diptera (Elouard, 2009). In 1989, Villemant reported the presence of four species of Hymenoptera antagonists of *O. trigotephras*: *Cotesia melanoscela* (Ratzeburg) and *Meteorus pulchricorni* (Wesmael), larval parasitoids (Bastaoui, 1983; Dahou, 1984), *Coccygomimus maraguesi* (Schmiedecknecht) and

[a] National Institute of Research in Rural Engineering, Water and Forests (INRGREF), Bp 10, 2080
[b] University of Sciences of Tunis, Tunisia
[*] **Corresponding author** (İletişim yazarı): sonia.hamamii@gmail.com
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Coccygomimus instigator (F.), pupal parasitoids (Villemant, 1989).

No work has been done on the auxiliary fauna of *O. trigotephras* in Tunisia and the Mediterranean basin. In this context, this work aims to study the importance of predation and parasitism of *O. trigotephras* and to better understand the relationship between pest/parasitoid and host plant in the North-east (Jebel Abderrahamane) and the North-west (Sejnane) of Tunisia in 2013.

2. Material and methods

2.1. Study area

This work took place in the north-eastern part of Tunisia (Jebel Abderrahmane, Cap Bon) and the north-western (Sejnane). One infested station in Sejnane (Barrage Ziatine) and four infested stations in Jebel Abderrahmane: Ftahiz, Delhiza and Guitoun (Tableau 1). The vegetation consists of Mediterranean maquis with shrubby vegetation, about 1-2m high. The predominant plant species are *Calicotome villosa* Poiret, *Cistus crispus* L., *Cistus monspeliensis* L, *Erica arborea* and *E. multiflora*, *P. lentiscus*, *Phillyrea media* L. and *Q. coccifera*.

Table 1. Study area

Stations	Altitude	Latitude (N)	Longitude (E)
Ftahiz	121m	36°.52’	10°.45’
Delhiza	401m	36°51'	10°47’
Guitoun	136m	36°,83’	10°,82’
Sejnane	48m	37°11′	9°11’

2.2. Sampling

A total of 30 egg masses were collected from each host species: *Q. coccifera* and *P. lentiscus* on each station and at the end of each generation during 2012 and 2013. Eggs masses were collected by cutting branches with scissors. In the laboratory, each egg mass was placed separately in a tube and was surveyed daily to follow-up the emergence of the natural enemies (parasitoids and predators) and to establish their density. The emerging insects were removed carefully using a brush of fine hair and after a relaxation time of 2 min in the refrigerator they were stored in an Eppendorf tube filled with ethanol (96%).

To identify viable, hatched, preyed, dried and unfertilized eggs we used the method described by Villemant (1993). Firstly, we removed the silky cocoon and then separate the eggs by brushing the egg mass inside a 1 mm mesh sieve to remove the scale layer that separate the eggs. Eggs were then placed in a 9 cm Petri dish and observed under a binocular microscope (Leica, S42). Total number of eggs per egg mass was counted as an estimate of the realized fecundity. Analysis of batches allowed the estimation of the average fertility of the female (the total number of eggs per batch), the rate of parasitism and predation for each batches during both spring and autumnal generations.

2.3. Statistical analysis

The statistical treatment of the data was performed using SPSS (version 17.0) software. For the analysis of female fecundity and the different categories of eggs ANOVA and Student-Newman-Keuls test were used to compare variables at 5%.

3. Results

3.1. Females fecundity

There is a high significant difference of fecundity between generations in each site (p <0.01). Female fecundity is higher in the SG than the AG (Figure 1). A high significant difference was observed between Ftahiz, Delhiza, Guitoun and Sejnane for the SG and the AG (p <0.01). Mean fecundity observed in Sejnane for both generations is more important than Jebel Abderrahmane (Figure 2).

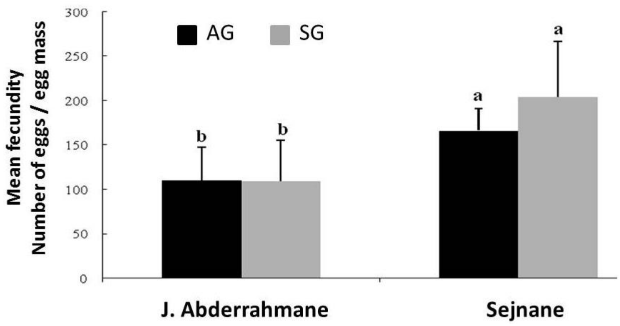

Figure 1. Mean fecundity of *O. trigotephras* for the two generations in both sites.
SG: spring generation; AG: autumnal generation

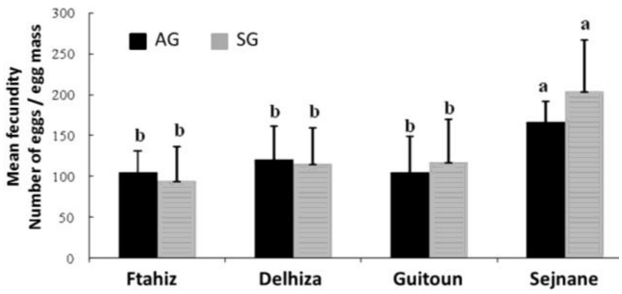

Figure 2. Mean fecundity of *O. trigotephras* for the two generations in the different stations
SG: spring generation; AG: autumnal generation

3.2. Egg mortality

Statistical analysis showed a significant difference (p<0.01) between hatched, predated and dried eggs in the two study sites and during the AG and the SG. No significant difference for the parasitized eggs (SG: p<0.73 and AG: p<0.68) and the unfertilized eggs (AG: p<0.193 and SG: p<0.71) between the two sites.

3.3. Parasitized and predated eggs

Ezzine et al. (not published) identified only one parasitoid of the genus *Aprostocetus* (Hymenoptera, Eulophidae) and only one predator, the moth *Coccidiphila rungsella* Nel & Brusseaux, 1997 (Cosmopterigidae) from eggs in 2005 and 2009. In this study, we confirmed the occurrence of both species for both spring and autumnal generations. *C. rungsella* was observed in Jebel Abderrahmane and in Sejnane, on *Q. coccifera* and *P. lentiscus* while *Aprostocetus* sp. was observed only in Jebel Abderrahmane, on the two host species.

Predation differed between generations (p<0.000), while parasitism did not (p=0.786). Between all the studied stations, predation and parasitism differed significantly (p<0.000). Although, between host plants, the difference was not significant with p=0.527 for predated eggs and p=0.338 for parasited eggs. Parasitism and predation in Jebel Abderrahmane were higher for both host plants in AG. For the SG, parasitism was important only on *P. lentiscus* (Table 2). However, in Sejnane parasitism was very low on both host plants for the AG, while for the SG, it was observed only on *Q. coccifera* (Table 2). In Ftahiz (AG), *Q. coccifera* was more parasitized than *P. lentiscus*. Whereas, predation was important on both host plants (Table 3). Although in SG, parasitoid and predator were observed only on *P. lentiscus* (Table 3). In Delhiza and Guitoun, parasitism was lower for both generations and both host plants. Yet, predation was important on both stations and host plants (Table 3).

4. Discussion

The difference of the female fecundity between the two generations can be due to the host plant and to the quality of the foliage of the host plant that can affect insect performance. *O.trigotephras* is a oak defoliator (Villemant & Fraval, 1993), young larvae (first and second instars larvae) of the SG feed on the fresh foliage of *Q. coccifera*. Older larvae feed on *P. lentiscus* and other shrub species (Ezzine *et al*, 2010). Young larvae of the AG feed on the fresh foliage of *E. arborea* and *P. lentiscus*. Older larvae feed on *Q. coccifera* (Ezzine *et al*, 2012). In 2005 and 2009, egg batches were found more frequently on *P. lentiscus*. Yet, larval survival, development and adult fecundity were higher when larvae could feed on *Q. coccifera* leaves (Ezzine et al., not published).

Parasitism and predation differs between the two generations, sites, stations and host plants. This difference is may be due the interaction between enemies, eggs and host plants. Ezzine et al. (2010) showed that there's a highly significant difference of parasitized eggs on different host plants: 1.2% on *Q. coccifera* and 0.2% *P. lentiscus*. In fact, some parasitoids or insects are able to attack their host plant at a specific stage of development thereof, usually during the bud break, so they must be present at the phonological stage of the host plant forward or lags behind its development, resulting in a reduced attack in duration or, in extreme cases, impossible (Marçais et al., 2000). While, when insects are specific to one or more host plants, they must develop a system that allows them to quickly find their site of feeding or breeding. At the location of their host plant, insects especially using vision, olfaction and taste for the oviposition (Claude Nicole, 2002). The epidemic development of many parasitoids is strongly influenced by the conditions of temperature and humidity, infection or multiplication (Marçais et al., 2000). The increase in temperature also plays on parasitoids and predators of processions pests (Mattson and Haack, 1987).Their thermal optimum may be different, global warming would sometimes be favorable to insect pests (Fleming and Volney, 1995), sometimes favorable to their natural enemies (Skirvin et al., 1997). The absence of *C. rungsella* in Guitoun can be explained by the difference of the altitude. It is likely that this predator prefers high altitudes (Delhiza) and the exposure to the sun and the sea (Sejnane). Qualitative and quantitative changes in host plants, climatic conditions for each station and even the biology of *O. trigotephras* can influence the success of the enemies. A study done in Morocco by Villemant and Fraval (1992) showed that climate and certain biological characteristics of the species (larval development, larval quiescence or prolonged imaginal) allow egg parasitoids to act during the egg stage of *Lymantria dispar*.

Natural enemy abundance differed between generations, sites and stations. These variations are due primarily to the nature of the host plant which is necessarily correlated to the climatic conditions. In this case, it is important to study the effects of climatic factors on the dynamics of the natural enemies in relation to the outbreak phase of the pest.

Table 2. Parasitism and predation between sites (mean ± SE). SG: spring generation; AG: autumnal generation

Generation	Eggs categories	Host plant	Sites	
			J. Abderrahmane	Sejnane
AG	predated eggs	*Pistacia lentiscus*	9,91±1,71	4,63±1,42
		Quercus coccifera	9,5±2,65	5,4±1,78
	parazited egg	*Pistacia lentiscus*	2,25±0,58	0,34±0,23
		Quercus coccifera	4,56±1,53	0,12±0,11
SG	predated eggs	*Pistacia lentiscus*	4,05±1,01	0
		Quercus coccifera	0,57±0,23	1,53±0,51
	parazited egg	*Pistacia lentiscus*	3,42±0,86	0
		Quercus coccifera	0,84±0,57	0,57±0,37

Table 3. Parasitism and predation among stations (mean ± SE). SG: spring generation; AG: autumnal generation

Generation	Egg categories	Host plant	Stations			
			Ftahiz	Delhiza	Guitoun	Sejnane
AG	predated eggs	*Pistacia lentiscus*	13,42±1,10	9,18±2,35	7,13±1,68	4,63±1,42
		Quercus coccifera	15,22±3,42	5,99±1,69	7,3±2,84	5,4±1,78
	parazited egg	*Pistacia lentiscus*	5,91±0,98	1,55±0,40	0,7±0,37	0,34±0,23
		Quercus coccifera	8,94±2,30	1,76±0,56	2,98±1,74	0,12±0,11
SG	predated eggs	*Pistacia lentiscus*	10,57±2,42	0,78±0,23	0,81±0,37	0
		Quercus coccifera	0	0	1,71±0,69	1,53±0,51
	parazited egg	*Pistacia lentiscus*	7,11±1,46	1,1±0,31	2,07±0,81	0
		Quercus coccifera	0	0	2,54±1,73	0,57±0,37

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