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Status, Distribution and Parasitism Rate of Olive Fruit Fly (*Bactrocera oleae*.Rossi) Natural Enemies in Lebanon

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Abstract

Olive tree is considered one of the most important cultivated crops in Lebanon. The cultivated area is mainly concentrated in the north and south Lebanon. Among the obstacles affecting olive cultivation is the olive fruit fly infestation which negatively alters the quality of olive oil. Olive fruit fly (*Bactrocera oleae*) is mainly controlled by chemical means. At the present time, no biological program against this pest is considered in Lebanon. The aim of this study is to investigate the status of olive fruit fly parasites in Lebanon. The study was conducted for one year in 28 orchards located in five regions of olive production. Fruit samples were collected weekly from June to October. A total of 23096 fruits was collected from all orchards. Eight parasites were detected. *Eupelmus urozonous* and *Opius concolor* were the most abundant parasites in most orchards, the others were restricted to specific locations. The only exception was observed at Abdeh site where most of the parasites were isolated. Parasitism rates ranged from 2.4 to 42.1 % and the average rate of all locations was 13.3%. The low parasitism rate recorded in this study is insufficient to control olive fruit fly. Introduction and rearing of natural enemies along with other management tactics may enhance the control of olive fruit fly infestation

Keyword: Olive tree, *B.oleae,E.urozonus,O.concolor*. Parasite, Parasitism rate.

1. Introduction

Olive tree is considered one of the most important cultivated crops in Lebanon. It represents about 43% of the cultivated area with fruit trees (FAO, 2012). The main cultivated area with olive trees is concentrated in the North and South Lebanon. In Mount Lebanon olive trees are mainly grown in Chouf region (FAO, 2012). The fruit infestation by *B.oleae* (Rossi) (Diptera: Tephritidae) is the main threat affecting olive cultivation in Lebanon. The adult females deposit their eggs (50-400 eggs in her lifetime) under the skin of the fruit(Daniela Merchini et al.,2017). The Larvae develop and feed on the pulp of the fruit. Infested fruits often drop prematurely. Secondary infestation of bacteria and fungi accumulated in the tunnels made by the larva inside the fruits decreases oil quality (F. G. Zalom, et.al,2003).

Chemical pesticide is the main option used to control olive fruit fly. Other option like attract and kill devices that minimize the impact of full cover spraying may offer an alternative management method to the use of synthetic pesticides (Iannotta N. et al,. 2010). Biological programs are scarce despite the existing of some parasites of *B. oleae* . In Lebanon, little information about parasitoid fauna is reported .Currently, no biological program against this pest is considered in Lebanon. E. Mechelany (1969) reported in his study five parasitoids(*Opius cocolor* Szepl., *Eupelmus Urozonus* Dalm., *Cyrtoptyx dacicida* Masi., *Pnigalio mediterraneus* Del., *Eurytoma rosa* (Nees.)) to be present in Lebanon. The first two of them are the most common.

O. Concolor and other parasitoids of the Bracinoid family are reported to be native to Africa (Biliotti, E. 1961, Cameron, E. 1941, Fischer, M. 1963). They were introduced to Europe (Fischer, M. 1963, Tobias, V. I. 1977, Monaco R. 1978), Jordan (Mustafa T. M. and Al-Zaghal K. 1987), Lebanon (Brnetic D. and Domazet D. 1981) and to other countries.

Four chalcidoid wasps were reported to be present in southern Europe *E. urozonus* (Hym.: Eupelmidae), *P. mediterraneus* (Hym.: Eulophidae), *Cyrtoptyx latipes* (Rondani) (Hym.: Pteromalidae) and *Eurytoma martellii* Domenichini (Hym.: Eurytomidae). Many of chalcidoid species are not specific to olive fruit fly and they attack unrelated hosts in several different insect orders mostly the family of tephritidae (Kim A. Hoelmer et al.,2011)

The objective of this study is to investigate the status, distribution and parasitism rate of olive fruit fly natural enemies in Lebanon for potential developing of future bio-control programs.

2.Methods and Material

The study was carried out for one year (2014) in 28 olive orchards located in five regions(Tyre,Nabatieh,Saida-Jizzin,Chouf,North). These orchards are mainly planted with local Baladi variety except for Abdeh orchard(North) which was planted with various local and imported varieties. The orchards are located along the littoral and up 850 m above sea level. The fruit sampling began in June and continued until the end of October .The fruits were kept in aerated transparent plastic boxes under ambient lab temperature for one month. The emergence of fruit fly and parasites were recorded every 2-3 days. Parasitism rate was calculated by the following formula: 100xP/(P+F),where (p) the total number of emerged parasites ;(F) the number of olive fruit flies. In addition to pheromone traps ,McPhail traps baited with a 3% of diammonium sulfate solution were placed in each orchard for monitoring the dynamics of olive fruit fly. The level of infestation by *B.oleae* was recorded on a sample of 100 fruits from each orchard at four dates. Parasite species were identified at the Lebanese agricultural research institute (entomology department). Statistical analysis was performed by using SAS for windows V8. A ttest was performed to discriminate between the means of captured flies on both pheromone and McPhail traps ,female and male of *B.oleae* and *O.concolor* and *E.urozonus*. Analysis of variance was performed to test the variation in the number of parasites and captured flies by locations and regions.

3. Results and Discussion

The captured flies on both McPhail and pheromone traps revealed a widespread of *B.oleae* throughout all selected regions(Table1). This can be explained by an assumption of the association of olive tree with *B. oleae* which was considered an indigenous to the Mediterranean basin (Kim A. Hoelmer et al.,2011). Captured flies on McPhail traps (M=32.8,SD=29.4) were significantly higher than on pheromone traps (M=13.2,SD=17.3), $F(1)=9.66$,$p<.0001$.The number of flies differed significantly between regions on both McPhail and pheromone traps respectively $F(5,284)=6.2$, $p<.0001$,$F(5,276)=12.6$, $p<.0001$.The highest number of flies was observed in Tyre ,Middle and coastal Chouf regions (Table1).Male and female flies were counted on McPhail traps and the results showed that male flies(M=19.5,SD=18.1) were significantly higher than female (M=11.5,SD=11.6),t(1)=2.3, $p<.0001$.

Table1.Average number of *B.oleae* adults captured on pheromone and McPhail traps from four regions during 2014.

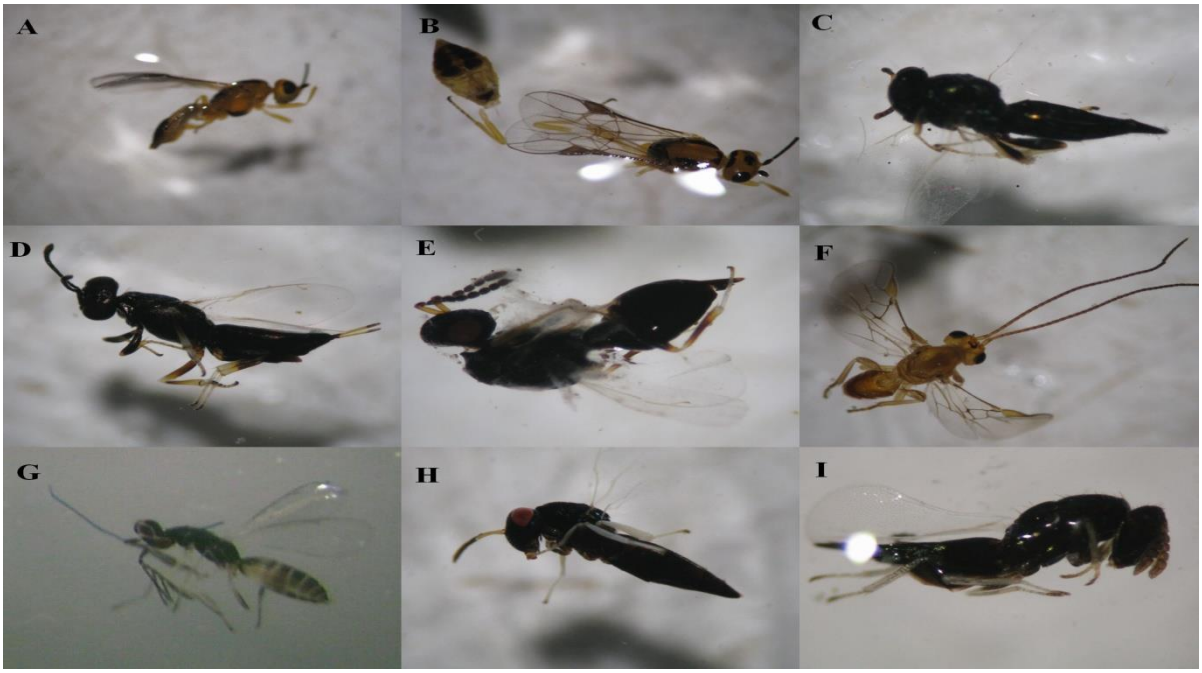
Region	Pheromone trap	McPhail trap (Diammonium sulphate,3%)		
		Total	male	female
Chouf				
High	7.7(12.0)b	22.9(23.6)b	11.9(11.7)d	8.0(8.1)bc
Middle	19.1(17.8)a	44.7(29.3)a	29.4(20.3)ab	13.6(12.4)b
Coastal	21.4(23.5)a	50(36.5)a	32.4(25.6)a	13.1(8.9)b
Jizzin-Saida	12.3(20.6)a	28.6(24.2)b	17.7(15)dc	11(9.8)bc
Nabatieh	7.7(5.2)b	17.4(12.1)b	11.2(8.1)d	6.2(4.5)c
Tyre	17(11.9)a	50.6(37.1)a	22.6(16)bc	28(22.4)a

Note. Columns with different letters are significantly different (Tukey's test, $p < 0.05$).

Eight parasitoids and one predator were recorded in this study: *Opius concolor* (Hymenoptera, Braconidae), *Eupelmus urozonus* (Hymenoptera Chalcidoidea), *Cyrtoptyx dacicida* (Hymenoptera, Pteromalidae), *Pnigalio mediterraneus* Del (Hymenoptera, Eulophidae), *Eurytoma spp*. (Hymenoptera, Eurytomidae), *Tetrastichus sp*. (Hymenoptera, Eulophidae), *Pteromalus* (Hymenoptera:Pteromalidae), *Bracon sp*.,(Hymenoptera,Braconidae) (Fig.1),and Prolasioptera berlesiana Paoli (Diptera, Cecidomyiidae). The first five were mentioned by Mechalany 1969 to be parasitoids of olive fruit fly in Lebanon.

Five parasitoids were isolated from olive fruit samples collected from Abdeh olive germplasm (North Lebanon)(Table2) where *E.urozonus* was the most common parasitoid. *Tetrastachus sp*.has been already reported to be a parasitoid of *B.oleae* in Jordan(Mustafa and Al- Zaghal 1987).*Tetrastachus sp*. has been reported to be present in Lebanon (M.Khouzami et al.,1996) but there is no information about this species as a parasitoid of *B.oleae.* It should

also be noted that there have been no reports about parasitism action of *Bracon sp*. and *Pterolamus sp* which were isolated from two olive fruit samples on olive fruit fly in Lebanon



Figrue1.Isolated parasitoids from olive fruit samples: (A) (B) *Bracon* sp.,(C)) *Cyrtoptyx dacicida*,(D) *Eupelmus urozonus*,(E) *Eurytoma* spp., (F) *O.concolor*,(G) *Pnigalio meditterranean* (H) *Pteromalus*, (I) *Tetrastichus* sp

Results showed a wide distribution of *E.urozonus* throughout all orchards. *E.urozonus* was caught from July till the mid of September (Table3).

In high elevation zone of Chouf region (Hasrout), *E.Urozonus* was recorded in the third week of October(Table3). It was also noticed a wide spread of *O.Concolor* in most orchards .The exception was observed in Sfarey (Jizzin region), Bazurieh and Aita al-Shaab(Tyre region) orchards where *O.concolor* was not recorded. Their activity was observed from-summer and continued until the third week of November (Table 3).Both species parasite third instar larvae , and can also parasite first and second instar. The other recorded parasitoids were rare and restricted to specific locations

Although the total number of *E.urozonus* was higher than the number of *O.concolor* , a ttest showed no significant difference between them (*E.urozonus* (*M*=7.3,*SD*=11.1), *O.concolor* (*M*=4.6,*SD*=4.4) and,(*F*(1)= -1.13,*p*=0.265)).

Table2: The distribution of 8 parasites isolated from olive fruit samples over the orchards of olive cultivation.

	O.Concolor	E.urozonus	C. dacicida	Pnigalio	*Eurytoma*	*Tetrastichus*	*Pteromalus*	Bracon
Chouf								
High elevation zone								
mmatour MLC1	+	+						
Atrin MLC2	+	+						
Baaklin MLC3	+	+						
Hasrout MLC4		+						
Moukhtara MLC5		+						
Middle elevation zone								
Debieh MLC1	+	+						
Chhim MLC2								
Joun MLC3		+						
Kfarfakoud MLC4	+	+						
Bourjein MLC5		+						
Coastal zone								
Aalman MLC1	+							
Rmeila MLC2	+	+	+					
Barja MLC3		+						+
Saida-Jizzin								
Ain Elmir1 SLC1	+	+						
Ain Elmir2 SLC2	+	+						
Baanoub SLC3	+	+			+	+		
Baisour SLC4	+							
Lebaa SLC5	+	+				+		
Mrah Hbas SLC6	+	+						
Salhieh SLC7	+	+						
Sfarei SLC8		+						
Nabatieh								
Arnoun NLC1	+	+	+					
Haboush NLC2	+	+	+					
Kfarrouman NLC3	+	+						
Tyre								
Bazourieh TLC1		+				+		
Aita Elshaab TLC2		+						
Bourj Alawei TLC3	+	+						
North								
Abdeh NLC1	+	+	+	+	+			

Table3.Dynamics and distribution of two parasitoids (O.concolor and E.urozonus) in Chouf, Saida-Jizzin, Nabatieh, Sour and Abdeh regions-2014

		July				August				September				October				November			
		1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
High elevation zone (Chouf region)																					
Ammatour	**O.concolor**																		+		
	E.urozonus												+	+							
Baaklin	**O.concolor**																				
	E.urozonus										+		+		+						
Etrin	**O.concolor**											+	+						+		
	E.urozonus										+		+	+	+						
Hasrout	**O.concolor**																		+		
	E.urozonus						+	+	+	+							+				
Moukhtara	**O.concolor**																				
	E.urozonus									+	+										
Middle elevation zone (Chouf region)																					
Bourjein	**O.concolor**														+				+		
	E.urozonus						+		+												
Debieh	**O.concolor**										+		+	+							
	E.urozonus					+	+		+	+	+			+							
Chhim	**O.concolor**																	+	+		
	E.urozonus																				
Joun	**O.concolor**																				
	E.urozonus						+	+													
Kfarfakoud	**O.concolor**										+			+							
	E.urozonus					+		+		+											
Coastal area(Chouf region)																					
Alman	**O.concolor**							+	+				+		+	+				+	
	E.urozonus					+	+		+		+										
Barja	**O.concolor**																			+	
	E.urozonus			+	+	+	+	+	+												
Rmeila	**O.concolor**	+					+								+						
	E.urozonus			+	+	+	+		+												
Saida- Jizzin region																					
Ain Elmir	**O. concolor**					+	+			+		+	+			+	+				
	E.urozonus												+								
Ain Elmir	**O. concolor**									+		+				+					
	E.urozonus					+				+											
Baanoub	**O. concolor**								+			+									
	E.urozonus						+			+											
Baisour	**O. concolor**									+						+	+			+	
	E.urozonus																				
Lebaa	**O. concolor**				+			+	+												
	E.urozonus				+		+	+	+												

Table3 cont.

		July				August				September				October				November			
		1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
Mrah Hbas	**O. concolor**						+		+					+						+	
	E.urozonus								+				+								
Salhieh	**O. concolor**									+			+								
	E.urozonus					+	+		+												
Sfarei	**O. concolor**																				
	E.urozonus									+											
Tyre region																					
Aita Elchaab	**O. concolor**																				
	E.urozonus														+						
Bazourieh	**O. concolor**																				
	E.urozonus											+									
Bourj Alawei	**O. concolor**																+				
	E.urozonus																				
Nabatieh region																					
Arnoun	**O. concolor**										+	+	+	+		+	+				
	E.urozonus					+		+				+	+	+							
Kfarrouman	**O. concolor**								+				+		+				+		
	E.urozonus								+						+						
Nabatieh	**O. concolor**										+		+	+	+				+		
	E.urozonus								+												
North region																					
Abdeh	**O. concolor**										+										
	E.urozonus			+	+	+	+	+			+										

In addition to these parasitoids species , *Chelonus blackburni* (Hymenoptera,Braconidae) (Fig.2) was also isolated from different olive fruit samples. The inspection of these samples revealed the wide spread of this species in four regions(Chouf,Jizzin,Nabatieh and Tyre regions) .They were found in 15 orchards with a total number of 59 adults .This wasp is a solitary endoparasite of Lepidopterous larva (Dennis S.Hill,1994).Based on this fact we assume that olive moth (*Prays oleae*) which belongs to the Lepidoptera family was parasited by *C.blackburni*. Future studies should be conducted to prove this assumption.

Figure2. *Chelonus blackburni* isolated from olive fruit samples.

2165 flies and 333 parasitoids were emerged from the 23096 collected olive fruits(Table4). The parasitism rate highly varied across all locations and ranged from 2.4% (Ammatour) to 42 %(Baanoub)(Table4).

Table4.Number of fruits, parasitoids ,emerged flies and parasitism rate recorded from four regions during 2014.

Region	No of fruits	No of flies emerged	No of parasitoids emerged	% of parasitism
Chouf				
High elevation zone				
Ammatour MLC1	1017	80	2	2.4
Atrin MLC2	1372	74	12	14
Baaklin MLC3	1096	64	5	7.2
Hasrout MLC4	1215	151	36	19.3
Moukhtara MLC5	780	68	6	8.1
Middle elevation zone				
Debieh MLC1	1215	143	24	14.4
Chhim MLC2	1145	87	2	2.2
Joun MLC3	200	11	2	15.4
Kfarfakoud MLC4	545	85	6	6.6
Bourjein MLC5	1024	121	11	8.3
Coastal zone				
Aalman MLC1	958	71	17	19.3
Rmeila MLC2	768	99	13	11.6
Barja MLC3	1110	57	13	20.3
Saida-Jezzin				
Ain Elmir1 SLC1	800	93	8	7.9
Ain Elmir2 SLC2	649	40	10	20
Baanoub SLC3	400	22	16	42.1
Baisour SLC4	765	66	10	13.2
Lebaa SLC5	1380	167	72	30.1
Mrah Hbas SLC6	762	92	7	7.1
Salhieh SLC7	675	42	10	19.2
Sfarei SLC8	630	14	1	6.7
Nabatieh				
Arnoun NLC1	848	92	16	14.8
Haboush NLC2	949	116	9	7.2
Kfarrouman NLC3	993	117	10	7.9
Tyre				
Bazourieh TLC1	600	59	2	3.3
Aita Elshaab TLC2	600	24	2	7.7
Bourj Alawei TLC3	600	110	11	9.1
	23096	**2165**	**333**	**13.3**

Parasitism rates in high and middle elevation zones of Chouf region were 10 and 8.9% respectively. In coastal area of Chouf region the P.rate reached 19%. Nearly the same rate was observed in Saida-Jizzin region.The average parasitism rate of all locations was 13.2 %(Table 4). These results agree with the results of Machelany and Abdelwali (1993) reporting in their works that parasitism by *P.concolor* reached peak levels of 12-16%. One of reasons for this low parasitism rate may be a lack of synchronization between the life cycles of the parasitoid and fly (Kyriaki Varikou et al., 2014).Fruit infestation by olive fruit fly did not differ significantly among regions (F (5, 82) =0.71, p=0.618, η^2=0.04) as well as among locations (F (21, 66) =0.57, p=0.913, η^2=0.15). A regression analysis was conducted to reveal the relationship between infestation and parasitism rate .The results showed that infestation did not predict significantly parasitism rate , β =.968, t(85) = 1.34, p = .1837, η^2=0.0093.

4.Conclusion

The results of this study showed a widespread of *O.concolor* and *E.urozonus* in nearly all locations. The other isolated parasitoids were rare and restricted to specific locations. In addition to the *B.oleae* parasitoids that were already mentioned by Mechalany 1969, *Tetrastachus sp. ,Pteromalus* sp,*and Bracon sp*.are the new parasitoid species attacking *B.oleae* in Lebanon. Despite small number of parasitoids found in this study, many of the natural enemies of *B.oleae* may still be undiscovered in Lebanon .The low parasitism rate recorded in this study is insufficient to control olive fruit fly. Introduction and rearing of natural enemies along with other management tactics may enhance the control of olive fruit fly infestation.

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