Boll. Zool. agr. Bachic.
Ser. II, 30 (2): 213-222

30 December 1998

V. CALECA, G. LO VERDE, S. BLANDO, V. LO VERDE

New data on the parasitism of citrus leafminer (*Phyllocnistis citrella* Stainton, Lep. Gracillariidae) in Sicily (*)

Abstract - From March to October 1998 the parasitism trend on citrus leafminer (CLM) was studied in two unsprayed orange groves of Agrigento province. In order to evaluate the parasitoid complex, samples were also collected, in the same period, in three other citrus groves. The leafminers of the spontaneous vegetation inside and near the orange groves were also investigated, with the aim of recording the Eulophid species known as parasitoids of CLM.
Because of the significant correlation resulted between the parasitism rate calculated as mature larvae and pupae of parasitoids compared with CLM pupae and the parasitism rate on L3, L4 and pupae, the first index was preferred, representing an easier way to assess the parasitism, especially when a large number of samples have been collected. During the spring flush CLM infestation was almost absent; in summer an increasing parasitism has been recorded in both orchards, reaching the maximum in September, 87.5% at Ribera and 24% at Menfi. A maximum mean value in this month was also recorded in the previous years. In the orange groves the total parasitism was 47.2% at Ribera and 9% at Menfi. In 1998 the parasitism due to of Eulophid parasitoids showed an increase (from 9.8% in 1995-97 to 18.1%), mainly due to the addition of *Semielacher petiolatus* (Girault) activity. This exotic species, although found only from August 1998, reached at Ribera interesting values of parasitism (83.3%). In 1998, as in the previous years, *Cirrospilus pictus* (Nees) was the most abundant parasitoid species, but in two citrus groves *S. petiolatus* was the main parasitoid. Nevertheless the parasitoid complex in Sicily was unable to influence noticeably the CLM infestation. Though the differences observed on the spontaneous vegetation inside and near the two orange groves, at the moment we have no evidences to state that the presence of alternative hosts of CLM Eulophid parasitoids on spontaneous plants could greatly influence the parasitism on *P. citrella. Phyllocnistis saligna* (Zeller), found infesting *Salix alba* L., is here recorded as another host of *Cirrospilus diallus* Walker; *Caloptilia stigmatella* (F.) on *Populus nigra* L. was found for the first time as host of *Pnigalio agraules* Walker.

(*) Research funded by Assessorato Regionale Agricoltura e Foreste, Regione Sicilia "progetto di lotta integrata" and by M.U.R.S.T. (60%) funds.

Riassunto - *Nuovi dati sulla parassitizzione di* Phyllocnistis citrella *Stainton (Lep. Gracillariidae) in Sicilia.*

L'andamento della parassitizzazione su *P. citrella* è stato osservato, da marzo ad ottobre 1998, in due aranceti della provincia di Agrigento in cui non sono stati effettuati trattamenti insetticidi; contestualmente si sono raccolti dati quindicinali sul germogliamento e sul livello d'infestazione (larve e crisalidi per foglia). Inoltre, si sono studiati i fillominatori della vegetazione spontanea presente all'interno e nei pressi degli aranceti per verificare la presenza degli Eulofidi parassitoidi di *P. citrella* su ospiti alternativi. Nello stesso periodo, sono stati effettuati campionamenti in altri tre agrumeti per studiarne il complesso parassitario. La vegetazione del germogliamento primaverile non è risultata infestata. In estate si è registrata una parassitizzazione crescente con un massimo in settembre, dell'87.5% a Ribera e del 24% a Menfi; i valori massimi di parassitizzazione si sono avuti in questo mese anche negli anni precedenti. Nell'aranceto di Ribera la parassitizzazione totale è risultata del 47.2%, a Menfi del 9%. Nel 1998 essa ha avuto un incremento (dal 9.8% nel 1995-97 al 18.1%), dovuto principalmente all'azione di *Semielacher petiolatus* (Girault), specie recentemente segnalata in Sicilia. Questo Eulofide, sebbene riscontrato soltanto dall'agosto 1998, ha raggiunto a Ribera livelli di parassitizzazione del 83.3%. Nel 1998, così come negli anni precedenti, *Cirrospilus pictus* (Nees) è stato il parassitoide più abbondante, anche se in due dei siti studiati *S. petiolatus* è risultato più frequente. Dai risultati conseguiti, il complesso parassitario attualmente presente in Sicilia non è stato in grado di influenzare notevolmente le infestazioni della fillominatrice degli agrumi. Nonostante le differenze osservate sulla vegetazione spontanea presente all'interno e nei pressi dei due aranceti, attualmente i risultati conseguiti non evidenziano una notevole influenza della presenza di ospiti alternativi dei parassitoidi sulla parassitizzazione di *P. citrella. Phyllocnistis saligna* (Zeller), fillominatore ritrovato su *Salix alba* L., è qui segnalato quale nuovo ospite di *Cirrospilus diallus* Walker; *Caloptilia stigmatella* (F.) su *Populus nigra* L. è stato ritrovato per la prima volta quale ospite di *Pnigalio agraules* Walker.

Key words: Eulophid wasps, *Phyllocnistis citrella,* parasitism trend, parasitism index, citrus, Sicily, *Semielacher petiolatus.*

INTRODUCTION

Schauff *et al.* (1998) reports several species belonging to 41 genera of Chalcidoidea parasitising the citrus leafminer (CLM) all over the world. 15 Eulophid species were until now recorded in Italy (Caleca & Lo Verde, 1998; Mineo *et al.*, 1998).

In addition to a previous research carried out from 1995 to 1997 (Caleca & Lo Verde, 1998), during 1998 the parasitism trend and the CLM parasitoid complex in some unsprayed Sicilian citrus groves were observed.

MATERIALS AND METHODS

The parasitism trend of CLM in two orange groves located at Ribera and at Menfi (both in Agrigento province), was studied from March to October 1998. The first orchard was along Verdura river, close to a riverine vegetation mostly represented by *Populus nigra* L., *Salix pedicellata* Desf., *S. alba* L. and *Fraxinus oxycarpa* Bieb.. The Menfi orchard was surrounded by windbreaks of *Olea europaea* L. and *Cupressus sempervirens* L., and by spontaneous hedgerows with *Rubus ulmifolius* Schott. as dominant species, present also as small plants under the citrus canopy. A drip irrigation system was used in both the orchards.

In order to monitor the flushing, in Menfi grove, five twigs per plant, about 30 cm long, were marked on 10 plants; the number of new shoots per twig was recorded every 15 days. To evaluate the infestation level, in each orchard two groups of 40-50 shoots from 5-10 contiguous trees were collected. From each group of shoots 52 leaves (26 leaves 1-3 cm long, 13 from 3 to 5 cm, and 13 longer than 5 cm) were examined counting larvae and pupae per leaf. The presence of pre-imaginal stages of parasitoids was also recorded on third instar larvae longer than 2 mm (L3), fourth instar larvae (L4), and pupae of CLM infesting all the collected shoots. Larvae and pupae of parasitoids were reared until adults emerged.

At the same time, samplings of the leafminers of the spontaneous vegetation inside and near Ribera orange grove were also collected, with the aim of recording the Eulophid species known as parasitoids of CLM on alternative hosts.

Other 13 samples were collected in two orchards consisting of *Citrus limon, C. sinensis* and *C. deliciosa* (Luparello, Parco d'Orleans) in Palermo province, and in a citrus nursery (Terme Vigliatore) in Messina province.

A mean value of CLM larvae and pupae per leaf was calculated among the three length classes of the leaves.

The parasitism rate was calculated in two different ways:

1) mature larvae and pupae of parasitoids compared with CLM pupae, taking into account the data from the leaves longer than 3 cm of all the collected shoots; this index, already calculated for CLM by Caleca & Lo Verde (1998), is similar to that used by Peña *et al.* (1996) for CLM and by Celli (1960, 1963) for other leafminers;

2) all pre-imaginal stages of parasitoids found on L3, L4 and pupae in a sample of an equal number of leaves (50-80) of each of the three up-mentioned length classes.

The correlation index r between the two parasitism rates was calculated for Ribera, Menfi, Terme Vigliatore citrus groves, and two lemon groves whose data are from Caleca & Lo Verde (1998). T-test was used for the significance. Samples consisting of less than 50 L3, L4 and 50 pupae were excluded from this analisys.

RESULTS AND DISCUSSION

In 1998 a total number of 1808 CLM pupae, 1116 L3, L4 and 586 pre-imaginal stages of parasitoids were collected.

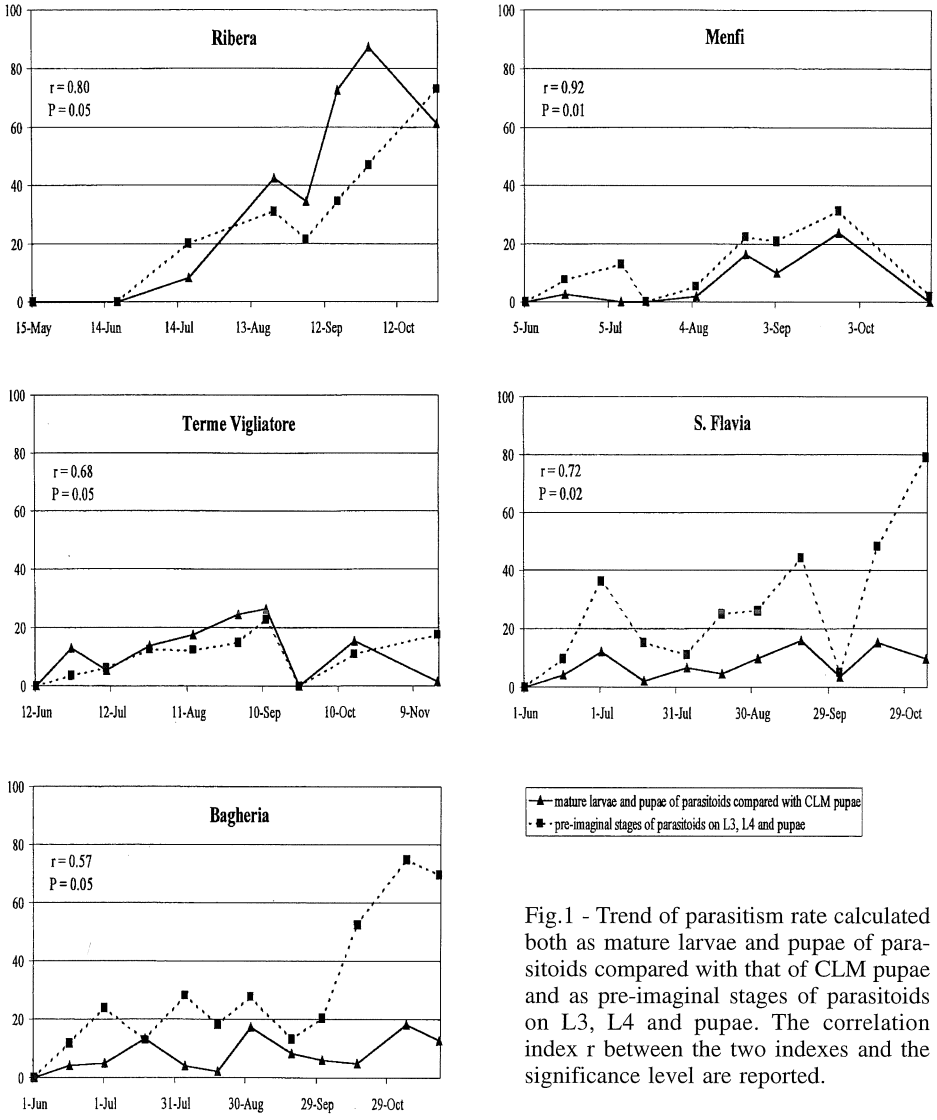

Fig.1 - Trend of parasitism rate calculated both as mature larvae and pupae of parasitoids compared with that of CLM pupae and as pre-imaginal stages of parasitoids on L3, L4 and pupae. The correlation index r between the two indexes and the significance level are reported.

The two parasitism rates resulted significantly correlated (fig. 1) at P= 0.05. Because of this we preferred to use the first index (mature larvae and pupae of parasitoids compared with CLM pupae) in the following analyses.

During the spring flush CLM infestation resulted absent. After the first recovery of parasitoids at Menfi on 19th of June, from July onwards an increasing parasitism rate was recorded, reaching a maximum of 87.5% at Ribera (fig. 2) and 24% at Menfi

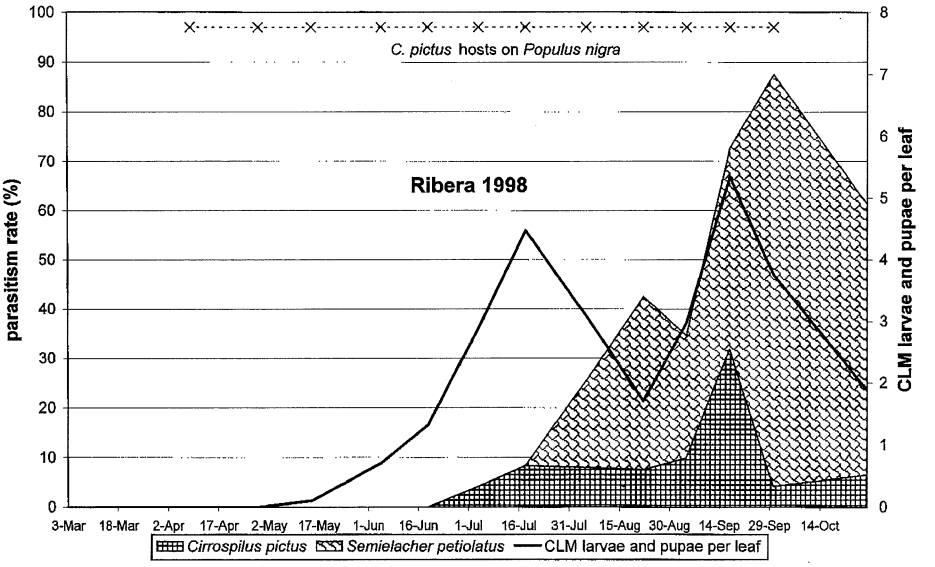

Fig. 2 - Trend of parasitism on *P. citrella* in the orange grove of Ribera from March to October 1998 compared with infestation and presence of alternative hosts on the spontaneous vegetation.

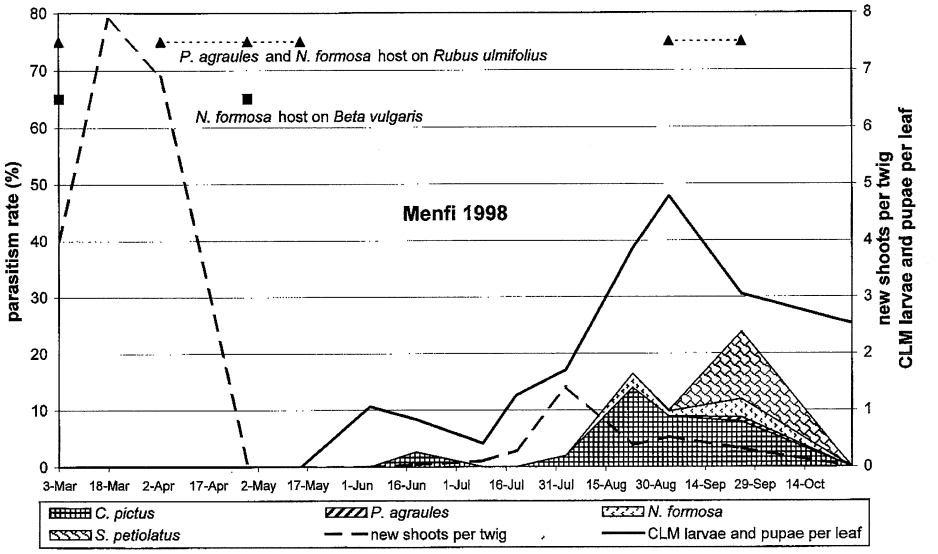

Fig. 3 - Trend of parasitism on *P. citrella* in the orange grove of Menfi from March to October 1998 compared with flushing, infestation, and presence of alternative hosts on the spontaneous vegetation. Data concerning spontaneous vegetation after Caleca *et al.* (1997) and Rizzo *et al.* (in press).

(fig. 3) in September; a maximum mean value in this month was also recorded in the previous years (fig. 5).

At Ribera two Eulophid species were found: *Cirrospilus pictus* (Nees) and *Semielacher petiolatus* (Girault) (fig. 2). At Menfi (fig. 3), besides these two species, *Neochrysocharis formosa* (Westwood) and *Pnigalio agraules* Walker also emerged.

S. petiolatus at its first recovery in Ribera on 22nd August was already the most abundant parasitoid (35%), reaching, at the end of September and in October, a parasitism rate of 83.3% (fig. 2).

The total parasitism in 1998 was 47.2% at Ribera (figs. 2, 4) and 9.0% at Menfi (figs. 3, 4), while in 1997 it was 17.6% and 4.8% respectively; these differences were

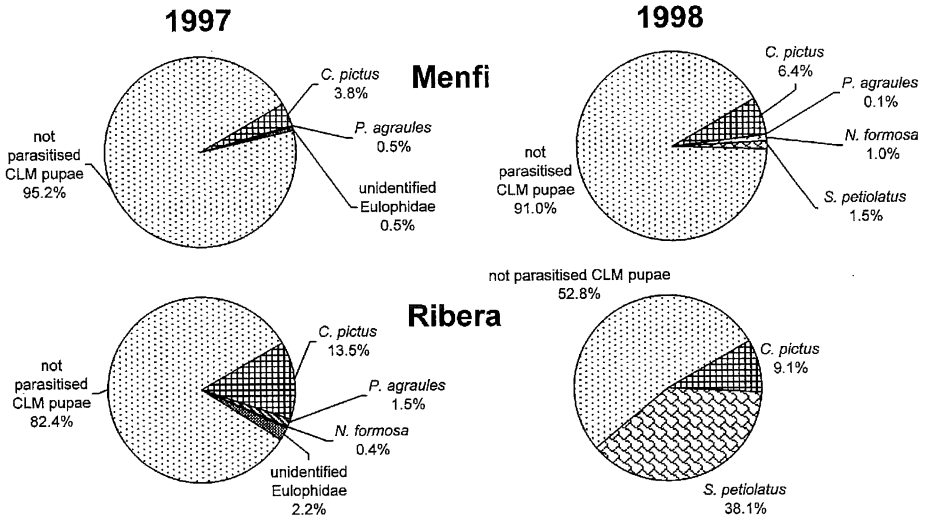

Fig. 4 - Parasitism rate recorded at Menfi and Ribera in 1997 (data from Caleca & Lo Verde 1998) and in 1998.

mainly due to the activity of *S. petiolatus* during 1998 (fig. 4). Moreover, during 1998 in the five sampled sites parasitoids reached 18.1% (fig. 6), while from 1995 to 1997, in the 12 sampled sites, they were 9.8% (fig. 7). Also in 1998 *C. pictus* was the most abundant species (49.0% of emerged parasitoids), but in two citrus groves (Ribera and Parco d'Orleans) *S. petiolatus* was the main parasitoid.

From the samplings of the riverine vegetation, some leafminers previously recorded as hosts of *C. pictus* (six species) and *P. agraules* (one species) in Ribera orchard (Caleca, 1998) were observed on *Populus nigra*. At least one of them was observed in each sampling until the end of September (fig. 2). *Stigmella trimaculella* (Haworth), *Phyllocnistis labyrinthella* (Bjerk.), *P. unipunctella* (Stainton) (Gracillariidae) were found parasitised by *C. pictus* also in this year, while *Caloptilia stigmatella* (F.) for

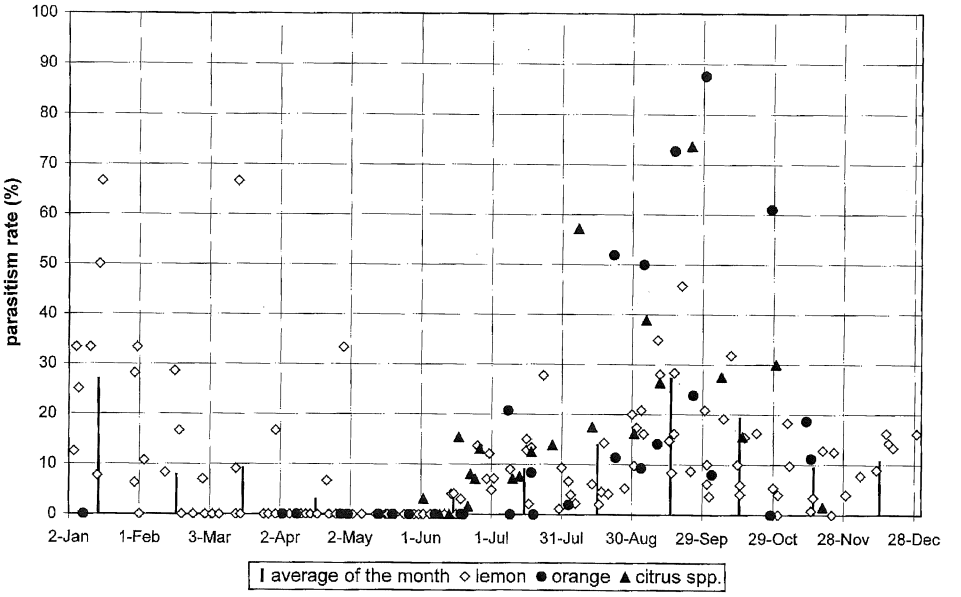

Fig. 5 - Seasonal parasitism rate of CLM in Sicilian citrus groves from October 1995 to October 1998 (171 samples).

the first time resulted parasitised by *P. agraules*. *Cirrospilus diallus* Walker was reared for the first time from *Phyllocnistis saligna* (Zeller) here collected on *Salix alba* from July to September.

From the spontaneous vegetation of Menfi orchard, from October 1996 to July 1998, Caleca *et al.* (1997) and Rizzo *et al.* (in press) reared *N. formosa* from the drosophilid *Scaptomyza* sp. on *Beta vulgaris* L., and *P. agraules, N. formosa*, and *Apotetrastichus postmarginalis* (Bouček), from the nepticulid *Stigmella aurella* F. on *R. ulmifolius* (fig. 3).

Though the abundance of alternative hosts of *C. pictus* at Ribera, its mean parasitism rate (9.1% Jun-Oct) was not so higher than Menfi's one (6.4%). In the latter orchard no host of *C. pictus* was found, but in spite of this, this Eulophid began to parasitise CLM two weeks before Ribera. The noticeable difference between the total parasitism of the two orange groves is mainly due to the unequal activity of *S. petiolatus,* which has not yet found on leafminers of spontaneous flora. Furthermore, *C. diallus* and *A. postmarginalis,* although found on spontaneous vegetation, were not recorded on CLM; a similar situation was observed for *P. agraules*, occurred at Menfi just once.

As regarding the parasitoid complex (tab. 1), besides the nine species collected up to 1997 (Caleca & Lo Verde, 1998), in 1998 *Asecodes erxias* (Walker) and *S. petiolatus* were also found.

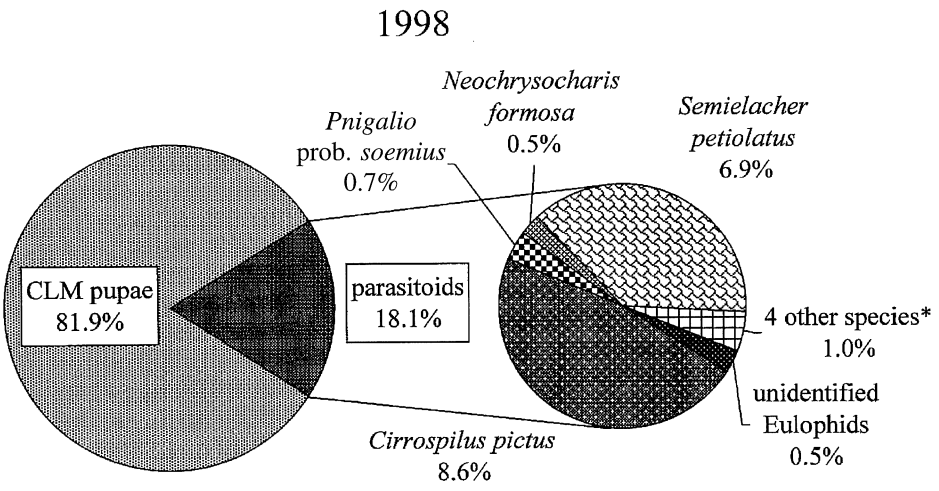

Fig. 6 - Parasitism rate recorded in 1998 (38 samples collected in five citrus groves).
*4 other species: *Cirrospilus diallus, Pnigalio agraules, Pnigalio* sp., *Asecodes erxias*

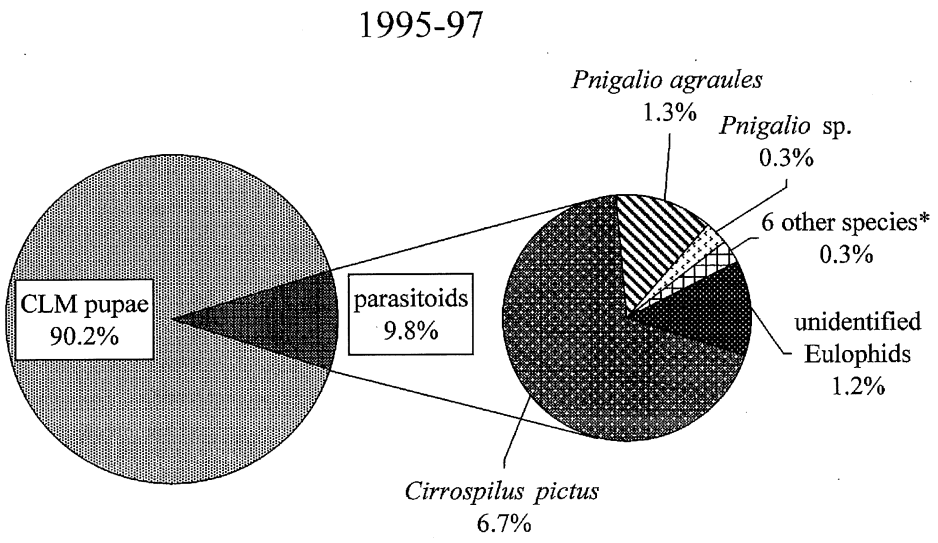

Fig. 7 - Parasitism rate recorded from October 1995 to December 1997 (133 samples collected in 12 citrus groves) (after Caleca & Lo Verde, 1998).
*6 other species: *Cirrospilus diallus, C.* sp. aff. *lyncus, Pnigalio* prob. *soemius, Ratzeburgiola incompleta, Apotetrastichus postmarginalis.*

Table 1 - Parasitism rate of Phyllocnistis citrella *in Sicilian citrus groves from October 1995 to October 1998.*

	Citrus limon	*Citrus* spp.	*C. sinensis*	Total	% among identified parasitoids
Sampling period	X.95 - XI.97	X.95-X.98	V.97-X.98	X.95 - X.98	
No. of sites	6	5	2	13	
No. of samples	118	23	30	171	
Examined specimens (CLM pupae + parasitoids)	10365	2078	2294	14737	
Cirrospilus pictus (Nees)	6.9	7.4	6.9	7.0	70.4
Cirrospilus diallus Walker	-	0.2	-	0.03	0.3
Cirrospilus sp. aff. *lyncus* Walker	0.01	-	-	0.01	0.1
Pnigalio agraules Walker	1.3	1.4	0.3	1.1	11.5
Pnigalio sp.	0.3	0.6	-	0.3	3.1
Pnigalio prob. *soemius* (Walker)	0.01	0.7	-	0.1	1.1
Asecodes erxias (Walker)	-	0.05	-	0.01	0.1
Neochrysocharis formosa (Westwood)	0.1	-	0.5	0.2	1.7
Apotetrastichus postmarginalis (Bouček)	0.04	0.1	-	0.04	0.4
Ratzeburgiola incompleta Bouček	0.01	0.6	-	0.1	1.0
Semielacher petiolatus (Girault)	-	0.9	5.8	1.0	10.4
Unidentified Eulophids	1.2	1.3	0.4	1.1	
Total % of parasitoids	9.9	13.3	13.9	11.0	
No. of species	8	9	4	11	

S. petiolatus was collected in all the five sites in August, September and October 1998, reaching 7,0% of parasitism in the year (fig. 6), and representing 38.9% of all the identified parasitoids in 1998.

CONSIDERATIONS

The parasitism rate calculated as mature larvae and pupae of parasitoids compared with CLM pupae can be used instead of the correlated parasitism rate on L3, L4 and pupae. It represents a easier way to assess the parasitism, especially when a large number of samples have been collected.

The results of the present study confirm that the parasitoid complex present now in Sicily was unable to influence noticeably the CLM infestation.

In 1998 the activity of Eulophid parasitoids shows a little increase, mainly owing to the addition of *Semielacher petiolatus*, an Australian species recently recovered in Sicily (Mineo *et al.*, 1998); this species, although found only from August 1998 onwards, reached in one orange grove interesting values of parasitism (83.3%, fig. 2).

Though the differences observed on the spontaneous vegetation inside and near the two orange groves, at the moment there are no evidences to state that the presence of alternative hosts of CLM Eulophid parasitoids on spontaneous vegetation could greatly influence the parasitism on *P. citrella.*

In 1998, as well as in 1995-97, *Cirrospilus pictus* has been the most abundant parasitoid species. The low parasitism level, recorded also in the previous years (Caleca & Lo Verde, 1998), and the low mortality due to this species also in the laboratory

(Lo Pinto *et al.*, 1998), could lead us to consider this parasitoid not promising for biological control programs involving rearing and releases.

ACKNOWLEDGEMENTS

We are grateful to Alessandro Fazzari, Nicolò Acquisto and Dario Butera for their help in collecting the material, and to Prof. Giovanni Mineo for the critical revision of the manuscript.

REFERENCES

CALECA V., 1998 - Parassitoidi riscontrati in Sicilia su fillominatori della flora ripariale limitrofa ad agrumeti e su *Phyllocnistis citrella* Stainton (Lepidoptera Gracillariidae). - Phytophaga (1996-97) 7: 45-55.

CALECA V., LO VERDE G., 1998 - Sul controllo naturale di *Phyllocnistis citrella* Stainton (Lepidoptera Gracillariidae) esercitato dai parassitoidi. - Phytophaga (1996-97) 7: 65-76.

CALECA V., RIZZO M. C., MASSA B., 1997 - Parassitoidi dei fillominatori della flora spontanea degli agrumeti della Sicilia.- Naturalista sicil. 22 (1-2):33-38.

CELLI G., 1960 - Ricerche sui parassiti di tre Microlepidotteri minatori delle foglie di Melo. - Boll. Ist. Ent. Univ. Bologna 24: 271-279.

CELLI G., 1963 - Contributi allo studio degli Imenotteri parassiti di Insetti minatori. - Boll. Ist. Ent. Univ. Bologna 26: 207-216.

LO PINTO M., SALERNO G., PERI E., 1998 - Azione parassitaria di *Cirrospilus pictus* (Nees) (Hymenoptera: Eulophidae) su *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillaridae). Nota preliminare. - Notiziario sulla protezione delle piante 8: 49-56.

MINEO G., CALECA V., MASSA B., 1998 - *Semielacher petiolatus* (Girault) (Hymenoptera Eulophidae), natural antagonist of *Phyllocnistis citrella* Stainton (Lepidoptera Gracillariidae), new for Italian entomofauna. - Naturalista sicil. 22(1-2): 3-6.

PEÑA J.E., DUNCAN R., BROWNING H., 1996 - Seasonal abundance of *Phyllocnistis citrella* (Lepidoptera: Gracillariidae) and its parasitoids in South Florida citrus. - Environ. Entomol. 25(3): 698-702.

RIZZO M. C., MASSA B., MINEO G., in press - Interrelations between the entomofauna of synanthropic vegetation and that one of citrus groves. – Proc. Symp. For Appl. Biol. Control in Medit. Countries, Cairo 25-29 Oct. 1998.

SCHAUFF M. E., LA SALLE J., WIJESEKARA G. A., 1998 - The genera of chalcid parasitoids (Hymenoptera: Chalcidoidea) of citrus leafminer *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae). Journal of Natural History 32 (7): 1001-1056.

DR. VIRGILIO CALECA, DR. GABRIELLA LO VERDE - Istituto di Entomologia agraria, Università degli Studi, Viale delle Scienze 13, 90128 Palermo, Italy

DR. SALVATORE BLANDO, Via Reina 25, 90011 Bagheria (PA), Italy

MISS VALENTINA LO VERDE, Viale Regione Siciliana 1245, 90129 Palermo, Italy

Accepted 30 November 1998