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Evaluation of the Egg Parasitoid *Edovum puttleri* Grissell (Hym. Eulophidae) for Biological Control of *Leptinotarsa decemlineata* (Say) (Col. Chrysomelidae) on Eggplant(1).

Introduction

The Colorado Potato Beetle (CPB), *Leptinotarsa decemlineata* (Say) (Col. Chrysomelidae), is found throughout Italy with the sole exception of Sardinia (Ugolini and Mancini, 1988). In the country's northern area it is the key pest of potato and eggplant (*Solanum melongena* L.).

The broad-spectrum insecticides (e.g. pyrethroids) employed against CPB on the latter crop make beneficial arthropod releases to control other eggplant pests impossible (Nicoli and Benuzzi, 1989). Any IPM strategy for eggplant must therefore be capable of controlling CPB by selective methods.

One of the most promising beneficial candidates for biocontrol of CPB is *Edovum puttleri* Grissell (Hym. Eulophidae), an egg parasitoid first discovered in Colombia in 1980 on *L. undecimlineata* (Stål) (Puttler and Long, 1983) and later in Mexico on *L. decemlineata* (Logan et al., 1987). The Colombian biotype of *E. puttleri* was imported into Italy from the United States in 1985 (Bin and Colazza, 1988) and has been reared on CPB eggs at the «Guido Grandi» Institute of Entomology in Bologna since February 1986 (Maini and Nicoli, 1990). *E. puttleri* was released in trials in the United States by Jansson et al. (1987), Lashomb et al. (1987), Williams (1987) and in Italy by Bin and Colazza (1988), Maini et al. (1988) and Pucci and Dominici (1988). All these trials involved adult parasitoid releases when females are at peak activity (Lashomb et al., 1987; Maini and Nicoli, 1990).The present paper reports the results of repeated *E. puttleri* releases against natural CPB infestations of eggplant grown both in plastic tunnel and in open field. It is to be noted that CPB can produce three generations per year in the trial area.

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Materials and methods

The trials were carried out in 1987 and 1988 at a farm in Forlì Province (northern Italy) where eggplant is regularly cultivated.

The local «Riminese» cultivar was planted at a density of about 9,600 plants/ha. The plants were grafted on nematode-resistant tomato and transplanted in plastic tunnel on April 29th 1987 and March 25th 1988 and in open fields on May 10th 1987 and April 30th 1988.

Each year *E. puttleri* was released against natural CPB infestations both in unheated PVC-covered greenhouse and in open field. The greenhouse (plastic tunnel) and the open field areas were each 240 m^2. The *E. puttleri* adults reared from CPB eggs collected on potato (Maini and Nicoli, 1990) (sex ratio ♀:♂ = 2-3:1) were released weekly 4 to 12 days after emergence. Between emergence and release, the parasitoids were kept in rearing units on honey, water and CPB egg masses.The number of parasitoid released depended on CPB infestation, i.e. on the number of egg masses found and on the number of reared *E. puttleri* available (figs. I,II). *E. puttleri* activity was monitored twice weekly by recording and dating by label all the CPB egg masses found on plants.

In 1987 the sampling was conducted by collecting all the egg masses a week after labelling, when most of the CPB larvae were hatched and the eggs had become unsuitable for *E. puttleri*. This was done to evaluate in laboratory the parasitization or the predation of each egg. Parasitized egg masses were considered as those from which one or more parasitoids developed, while the preyed upon egg masses were those killed but containing no *E. puttleri*.

In 1988 the labelled egg masses were left on the leaves instead of being collected. The number of eggs per mass was counted with a 10 x lens right after labelling. Hatched CPB eggs, predation, and *E. puttleri* parasitization and emergence were checked during the sampling no less than three weeks later. This to evidence any *E. puttleri* augmentative capability. The greenhouse whitefly, *Trialeurodes vaporariorum* (Westw.), and the red spider mite, *Tetranychus urticae* Koch, where controlled by releases of the parasitoid *Encarsia formosa* Gahan, and of the predatory mite *Phytoseiulus persimilis* Athias-Henriot (Nicoli and Benuzzi, 1989).

Statistical analysis

The data on the fate per sampling of CPB egg masses and CPB eggs within egg masses after *E. puttleri* releases were divided by frequency classes. The two distribution frequencies, i.e. plastic tunnel vs. open field, for each year were then compared using chi-square analysis. Mean differences of CPB egg fates within parasitized CPB egg masses were estimated using Normal Standard Deviation analysis.

Results and discussion

The data show that *E. puttleri* in both open field and plastic tunnel can find, prey upon and parasitize CPB egg masses. Besides *E. puttleri*, other common

predators, and sometimes CPB itself, can attack CPB eggs. When the eggs were preyed upon by entomophagous insects with sucking mouthparts, it was impossible to distinguish *E. puttleri* predation activity from that of such other predators as Chrysopidae and Nabidae, which were the most frequent ones found. The egg mass data thus include all such predation (figs. Ia, b; IIa, b).

In the 1987 plastic tunnel trial, parasitization started in June, about one month after the first egg mass was found. Subsequent *E. puttleri* activity continued up to the end of CPB egg laying (fig. Ia). In open field no eggs of CPB overwintering generation were found; *E. puttleri* was thus released only in early July and continued its activity, as in plastic tunnel, to early September (fig. Ib). Table 1 shows the fates of CPB egg masses recorded on plants; figure I (c, d) and tables 2 and 3 include the fates of CPB eggs collected one week after labelling for laboratory inspection.

In the 1988 trials *E. puttleri* parasitization of the egg masses laid by overwintering CPB was low in both tunnel and open field (fig. II a, b). These findings may be due to an environment generally unfavourable to *E. puttleri*, i.e. low temperatures and scarce sources of carbohydrates (Idoine and Ferro, 1988). The parasitoid's activity was far superior on the CPB egg masses laid by the subsequent generations, especially in tunnel throughout August (fig. IIa). Significant differences were found between the distribution of CPB egg mass parasitization in tunnel vs. open field (chi-square=12.13; $P<0.01$). This result was not observed during the first trial year, probably because the collection of all the parasitized egg masses prevented the augmentative capability of the parasitoid. The distribution of the parasitized CPB eggs was also higher in tunnel than in open field (chi-square=10.31; $P<0.05$) (figs. II c, d; tabs. 1, 2). Considering

Tab. 1 - Fates of CPB egg masses after the releases of *Edovum puttleri* in commercial eggplant in plastic tunnel and open field. n.s., not significantly different, $P > 0.05$; **, $P < 0.01$.

Variable	1987			1988		
	Plastic tunnel	Open field		Plastic tunnel	Open field	
No. of egg masses sampled	429	588	–	568	460	–
No. of egg masses parasitized	206	175	n.s.	345	162	**
(%)	(48.02)	(29.76)		(60.74)	(35.22)	
No. of egg masses preyed upon by other predators (%)	84	141	n.s.	78	101	n.s.
	(19.58)	(23.98)		(13.73)	(21.95)	
No. of egg masses hatched or partially preyed upon (%)	139	272	n.s.	145	197	n.s.
	(32.40)	(46.26)		(25.53)	(42.83)	
egg masses left on plants with wasp emergence (%)	–	–		(77.67)	(79.26)	

Tab. 2 - Fates of CPB eggs observed after releases of *Edovum puttleri* in commercial eggplant in plastic tunnel and open field. n.s., not significantly different, $P > 0.05$; *, $P < 0.05$.

Variable	1987 Plastic tunnel	1987 Open field		1988 Plastic tunnel	1988 Open field	
No. of eggs observed	7519	7977	–	20069	15864	–
No. of parasitized eggs	2725	2493	n.s.	4416	1632	*
No. of preyed upon eggs within parasitized egg masses	3708	3668	n.s.	7621	4093	n.s.
No. of preyed upon eggs within unparasitized egg masses	1028	1625	n.s.	4825	5941	n.s.
No. of eggs hatched	58	191	n.s.	3207	4198	n.s.

Tab. 3 - Fates of CPB eggs within egg masses with some eggs that were parasitized by *Edovum puttleri* released in commercial eggplant in plastic tunnel and open field. Means (± S.D.) are not significantly different, n.s., $P > 0.05$; *, $P < 0.05$.

Variable	1987 Plastic tunnel	1987 Open field		1988 Plastic tunnel	1988 Open field	
No. of eggs per egg mass	31.37±14.97	34.50±15.00	n.s.	35.71±15.21	36.59±16.41	n.s
No. of parasitized eggs	13.23±10.39	13.09± 9.12	n.s.	12.80± 8.80	10.08± 8.43	*
No. of preyed upon eggs	18.00±12.82	20.96±13.06	n.s.	22.09±12.98	25.27±14.43	n.s.
No. of eggs hatched	0.14± 0.64	0.45± 2.54	n.s.	0.82± 2.82	1.24± 2.96	n.s.
% of egg mortality	99.55	98.69	–	97.70	96.61	–
% of eggs parasitized	42.17	37.94	–	35.84	27.55	–
% of eggs preyed upon	57.38	60.75	–	61.86	69.06	–

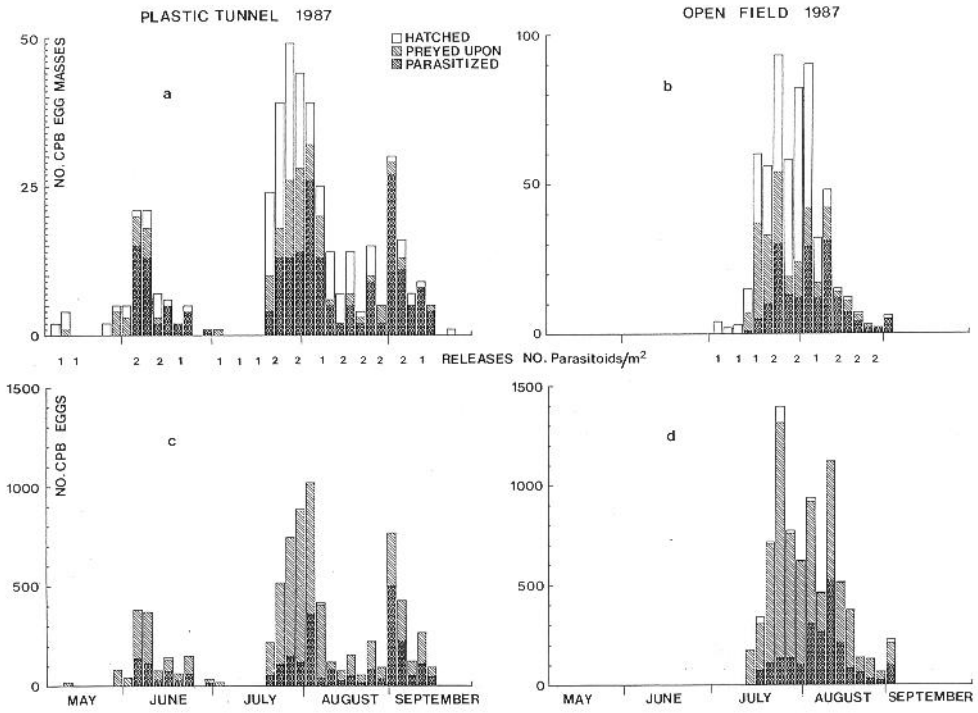

Fig. 1 - Seasonal trends of CPB egg masses and CPB eggs fates after *Edovum puttleri* releases in commercial eggplant.

the within-egg mass parasitism the mean of parasitized eggs was higher in tunnel than in open field ($d=3.26$; $P<0.002$) (tab. 3).

In 1988 many *E. puttleri* adults were also found to have emerged from the egg masses left on plants (fig. III). While it was impossible to distinguish their activity from that of the mass-reared *E. puttleri*, this fact provides a basis for the «seasonal inoculative releases» method (van Lenteren, 1986). Moreover, trials on organic farms in northern Italy showed that a single, initial-season release of *E. puttleri* resulted in parasitized egg masses even at the end of CPB egg laying (unpublished data). These data support the conclusion previously put forth by Lashomb et al. (1987) regarding the seasonal reproductive capability of *E. puttleri* in New Jersey eggplant fields.

The emergence of *E. puttleri* from egg masses that were left on plants and observed at least three weeks after labelling was 77.67% in tunnel and 79.26% in open field, with at least one *E. puttleri* adult emerged (29.04% labelled egg masses in tunnel and 42.66% in open field were lost). These field data show that *E. puttleri* is a viable biological tool for CPB control of field and protected eggplant crop in northern Italy. Research on *E. puttleri* release rates and on integrating other biological control techniques, e.g. sprays of *Bacillus thuringiensis* Berl. subsp. *tenebrionis*, are currently underway. Besides further assess-

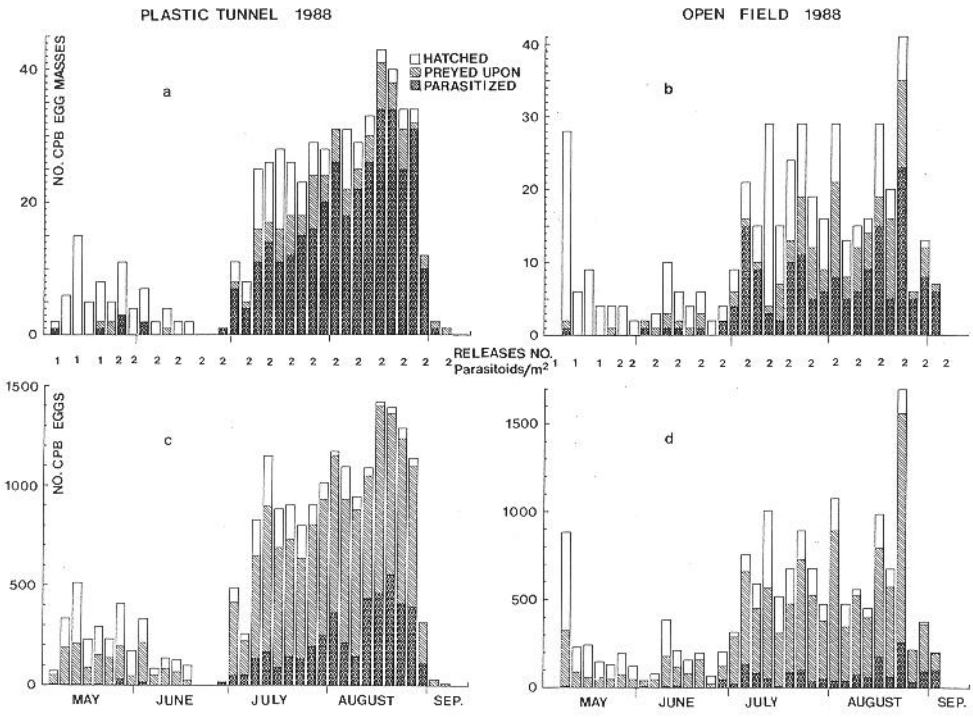

Fig. II - Seasonal trends of CPB egg masses and CPB eggs fates after *Edovum puttleri* releases in commercial eggplant.

ment of release techniques, i.e. pupae rather than adults, are also warranted. Pupae releases would facilitate storage and delivery of the reared parasitoid, these latter factors being very important for successful biological control.

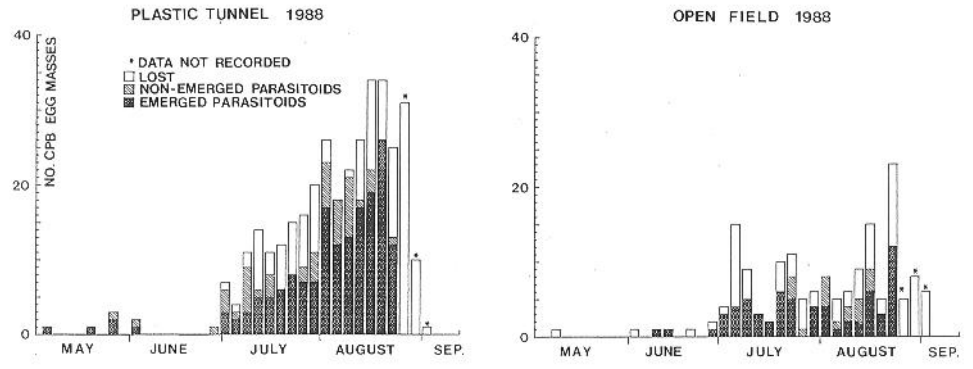

Fig. III - Seasonal emergence of *Edovum puttleri* adults from CPB egg masses.

SUMMARY

The Colombian biotype of *Edovum puttleri* Grissell (Hym. Eulophidae) was tested for two years against the Colorado Potato Beetle (CPB) *Leptinotarsa decemlineata* (Say) (Col. Chrysomelidae) on eggplant grown in plastic tunnels and open field. *E. puttleri* adults were released (1 or 2 per m^2) weekly 4-12 days after emergence throughout the CPB egg laying period. *E. puttleri* activity was monitored twice weekly by recording and dating by label all the CPB egg masses found on plants. In 1987 the sampling was conducted before adult emergence by collecting the CPB-labelled egg masses to evaluate in laboratory *E. puttleri* activity: parasitized and preyed upon eggs within egg masses. The percentage of egg masses attacked by the wasp was 48.02% in tunnel and 29.76% in open field. Of the egg masses exploited by *E. puttleri* the percentage of parasitized and preyed upon eggs was respectively 42.17 and 57.38% in tunnel and 37.94 and 60.75% in open field. In 1988 the labelled egg masses were left on the leaves to evidence *E. puttleri* augmentative capability. The percentage of egg masses parasitized was 60.74% and 35.22% in tunnel and open field respectively, and within egg masses *E. puttleri* parasitized 35.84% eggs in tunnel and 27.55% eggs in open field and killed an additional 61.86% and 69.06% eggs respectively by predation. The percentage of egg masses left on the leaves from which adults had emerged was 77.67% in tunnel and 79.26% in open field. The data show that *E. puttleri* could be a biological tool for CPB control of field and protected eggplant crops. The best results were achieved against the second and third generations.

Valutazione dell'attività di *Edovum puttleri* Grissell (Hym. Eulophidae) nei confronti di *Leptinotarsa decemlineata* (Say) (Col. Chrysomelidae) su melanzana.

RIASSUNTO

Nel 1987 e 1988, il biotipo colombiano di *Edovum puttleri* Grissell (Hym. Eulophidae) è stato provato contro *Leptinotarsa decemlineata* (Say) (Col. Chrysomelidae) su melanzana sia in tunnel plastici che in pieno campo. Durante il periodo di ovideposizione di *L. decemlineata* sono stati lanciati settimanalmente adulti di *E. puttleri*, di età compresa tra 4 e 12 giorni (1-2 per m^2). I campionamenti sono stati effettuati 2 volte alla settimana, contrassegnando e datando singolarmente tutte le ovature trovate sulla vegetazione. Per rilevare la parassitizzazione e la predazione, nel 1987, le ovature sono state portate in laboratorio, dopo una settimana dal ritrovamento. La percentuale di ovature attaccate da *E. puttleri* è stata pari al 48,02% in tunnel e al 29,76% in pieno campo. La percentuale delle uova parassitizzate e predate, relativamente alle ovature visitate da *E. puttleri*, è stata pari rispettivamente al 42,17 e 57,38% in tunnel e 37,94 e 60,75% in pieno campo. Nel 1988, le ovature contrassegnate sono state invece lasciate sulle piante, per valutare la capacità accrescitiva del parassitoide. Le ovature parassitizzate sono state rispettivamente il 60,74% in tunnel e 35,22% in pieno campo. Nelle ovature visitate da *E. puttleri*, la parassitizzazione e la predazione delle uova sono risultate rispettivamente il 35,84% e 61,86% in tunnel e il 27,55% e 69,06% in pieno campo. *E. puttleri* è sfarfallato dal 77,67% delle ovature lasciate in tunnel e dal 79,26% in pieno campo. *E. puttleri* è quindi un candidato per la lotta biologica contro *L. decemlineata* sulla melanzana e i migliori risultati sono stati finora raggiunti contro la seconda e terza generazione del fitofago.

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