8.　Parasitology

The diversity and specificity of parasitoids attacking Neotropical tortoise beetles (Chrysomelidae, Cassidinae)

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Abstract. Tortoise beetles have numerous adaptations to keep enemies at bay – including tightly-aggregated larvae that move synchronously about the food plant, construction of predator-deterring exuvio-fecal shields, maternal guarding of immatures, and adults that pull their carapace flush to the leaf to escape enemies. Despite these and other adaptations this subfamily of Chrysomelidae has been regarded as one of the most heavily parasitized. To better describe the impact and diversity of the parasitoid community which successfully evades these defenses we collected and reared the immature and adult stages of 47 species of Panamanian Cassidinae obtaining at least 41 species of parasitoids. Over half of the species obtained (26) were egg parasitoids (Eulophidae, Entedoninae), 20 of those *Emersonella* species, 13 undescribed at the time of the study. Phoresy was confirmed in at least six *Emersonella* species, two of which emerged from the eggs of 11 and 13 different host species. Nevertheless, the majority of Eulophidae species (15 of 26) were reared from a single host. Additionally, five species of Chalcidae, eight species of Tachinidae, two Nematomorpha and the lepidopteran, *Schacontia* sp. (Crambidae) were obtained from rearings of larvae, pupae and adults. One tachinid species (*Eucelatoria* sp.) infected the larval stage of *Chelymorpha alternans*, and was found in the abdomens of 27.6 percent of dissected adults.

Keywords. Cassidinae, Eulophidae, Tachinidae, parasitoids, parasitism.

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1.　　Introduction

A diverse array of predators, parasitoids and pathogens diminish the survival of phytophagous insects and thereby influence the dynamics of terrestrial ecosystems (LaSalle & Gauld 1991; Connor & Tavener 1997; Hawkins *et al.* 1997). Yet, the importance of parasitoids as a mortality source in key arthropod herbivore groups in the Tropics remains under-investigated.

'Tortoise beetles' (Cassidinae, *sensu stricto*; *i.e.* not including 'hispine beetles') represent a large subfamily (> 3000 species) of leaf beetles distributed world-wide, reaching greatest diversity in tropical latitudes (Windsor 1987, Jolivet & Hawkeswood 1995). Panamanian Cassidinae are associated with eight families of forest-edge and early successional herbaceous and woody host plants (Windsor *et al.* 1992), a notably small fraction of the 224 families of vascular plants available on the Isthmus (Correa *et al.* 2005). Five of these dicot families; the Convolvulaceae, Asteraceae, Bignoniaceae, Solanaceae and Boraginaceae, together host 88% of the 72 Panamanian tortoise beetle species whose host plants have been well established. The Convolvulaceae, host at least 27 species, 38% of the cassidine fauna. These same patterns were evident in the host plant records of 170 Cassidinae species from throughout the Neotropics (Buzzi 1994). Nearly all cassidines feed on hosts within a single plant genus and thus their feeding habits can be properly termed 'narrowly oliphagous'. The only exception known in the Panamanian fauna is *Polychalma multicava* (Latreille) which feeds on host plants in two families, various *Cordia* species (Boraginaceae) and *Helicteres guazumifolia* Kunth (Sterculiaceae). The dietary conservatism of tortoise beetles plus the exposed nature of their immature stages may make them more predictable and exposed targets for predators and parasitoids than other chrysomelid subfamilies (Cox 1994).

The Cassidinae have evolved numerous behavioral and morphological adaptations to cope with predators. These include broad, explanate margins of the pronotum and elytra which may function to prevent haustellate predators from piercing the body with an ovipositor or extended mouthparts. Additionally, larvae of most species of Cassidinae bear 'exuvial' or 'exuvio-fecal' shields which may serve as a mechanical or chemical barrier against predators (Olmstead 1996; Olmstead & Denno 1993; Eisner 1967; Vencl *et al.* 1999; Gomez 1999). Further, the resting larvae of some species gather into a compact circle ('cycloalexy') in which either the heads or the abdomens are positioned outward, forming a defensive phalanx with shields waved synchronously when disturbed (Vasconcellos & Jolivet 1994). In other species, larvae minimize their time on exposed peripheral foliage by traveling as a group toward the base of the plant where they reside motionless for hours before ascending again to feed (examples in Panama include *Polychalma multicava* and *Stolas ephippium* (Lichtenstein)). Some Cassidinae species attach excrement to eggs or enclose their clumped eggs in ribbons and folds of colleterial gland secretion (Becker & Frieiro Costa 1987; Muir & Sharp 1904; Hilker 1994). Females in some genera of Cassidinae (*Eugenysa, Acromis, Paraselenis, Omaspides*)

actively guard their eggs, larvae and pupae, thereby protecting them against predators and parasitoids (Windsor 1987; Windsor & Choe 1994). Since the majority of Cassidinae feed on host-plants with noxious phytochemicals (Olmstead 1996), including species in the Convolvulaceae (Blum 1994), it is likely that many Cassidinae obtain chemical defenses or their precursors from the secondary metabolites of the plants on which they feed. Indeed, Vencl & Morton (1999) working with the Solanum-feeding *Helocassis clavata* (F.) and Vencl *et al.* (2005) working with Convolvulaceae-feeding *Chelymorpha alternans* Boheman larvae have elegantly demonstrated the repellant properties of intact fecal shields relative to no shields or shields which have been leeched with polar and nonpolar solvents. Thus while Cassidinae appear to have many defensive adaptations, they also appear to have many enemies and in some cases their defenses may open the portals for specialized predators and parasitoids.

Following an exhaustive review of the literature on the Hymenoptera and Diptera attacking Chrysomelidae, Cox (1994) concluded that "The Cassidinae are the most frequently parasitized subfamily". Ten families of Hymenoptera and three subfamilies of Tachinidae were listed as parasitizing Cassidinae. The next most parasitized subfamily using these criteria were hispine beetles with nine parasitizing Hymenoptera families, but no parasitizing Tachinidae. While the results of Cox's literature survey could reflect differences in the sampling efforts of the various studies, his compilation remains our best indicator of the ecological importance of parasitoids across chrysomelid subfamilies. The number of reports which gauge the impact of parasitoids on Tropical Cassidinae are relatively few. Becker & Frieiro Costa (1987, 1988) found the eulophid, *Emersonella oecia* De Santis, accounted for the loss of 10-46% (average = 35.1%) of *Gratiana spadicea* Klug eggs in Southern Brazil, with larger oothecae being more highly parasitized and eggs in the inner core being not parasitized, probably because they were beyond the reach of parasitoid ovipositors. An even more important mortality source on the same oethecae was the mirid bug, *Tupiocoris cincticornis* Carvahlo, which sucked-dry 39.1% of eggs. *Emersonella neivipes* Girault destroyed 74-100% (average = 86.4) of the eggs of a *Stolas* sp. near Manaus, Brazil (Carroll 1978). A eulophid wasp (*Emersonella pubipennis* Hansson) accounted for the loss of 5, 26 and 35% of *Acromis sparsa* Boheman eggs, depending on whether oviposition sites occurred in forest, intermediate or open sites, respectively (Windsor 1987). One or more parasitoids eclosed from 77% of all egg masses in that study. In the Asian tropics, detailed field studies by Nakamura & Abbas (1987, 1989) documented the overall loss of 27.8% of *Aspidimorpha sanctaecrucis* (F.) eggs to *Cassidocida aspidimorphae* Crawford (Tetracampidae) and 39.8% of *A. miliaris* (F.) eggs to *Tetrastichus* sp. (Eulophidae), with mortality fluctuating seasonally between 40-60% and 10-80%, respectively, in the two species. Below we attempt to integrate both field notes taken over several decades and the results of a recent intensive study of parasitoids reared from 47 species of Panamanian tortoise beetles.

2.   Materials, methods and collecting sites

Systematic collections of Cassidinae eggs, larvae and adults by MC were made in the 'Parque Metropolitano' on the northern edge of Panama-City (elev. 40 m), 'Parque Soberania' near Gamboa (elev. 50 m) 20 km N of Panama City, and in the 'Parque Altos de Cerro Campana' approximately 50 km SW of Panama City (elev. 700-900 m). Collections were made at weekly intervals during both early and late wet season conditions (2 November to 24 December 2000, 2 May to 30 June 2001, and 3 May to 2 July 2002). Additionally, larvae were collected on three occasions at Chiriqui Grande (Bocas del Toro Province, elev. 20 m), Remedios (Chiriqui province, elev. 30 m) and between kms 8 and 9 along the El Llano-Carti Road (Panama Province, elev. 400 m). Collecting was largely restricted to Cassidinae species feeding on the genera *Ipomoea*, *Merremia* and *Bonamia* (Convolvulaceae), however occasional samples were made of species feeding on Boraginaceae (*Cordia alliodora* (Ruiz & Pav.) Oken), Asteraceae (*Mikania micrantha* Kunth and *M. guaco* Bonpl.) and Heliconiaceae (*Heliconia* sp.). These data were augmented by opportunistic collections made by DW and other researchers at these and nearby collecting sites in Panama.

Because eggs were infrequently encountered in the field and their identification often problematic, we enclosed beetles in mesh bags placed around aerial parts of the host-plant, one species per bag. The black organza which was used for bag construction was sufficiently coarse (2 × 2 mm) that egg parasitoids could easily enter to parasitize eggs but not sufficiently wide so that beetles could escape. Adult beetles were kept in the bags in the field for a sampling period of one week. Thus, at the end of the sampling period a collection of eggs was obtained with exposure to parasitoids varying between 0 and 7 days, a considerably shorter average exposure time (3.5 days) than eggs would have had under totally natural conditions. For this reason, the bagging study is likely to have underestimated the actual percentages of eggs parasitized. However, the bagging technique and its limitations are likely to apply nearly equally to all species in the study and therefore are useful in interspecific comparisons and the calculation of minimum percentages lost to parasitoids.

The number of bags set up for each species and the number of Cassidinae inserted into the bags varied depending on the abundance of the species at each site. In general, rarely encountered beetle species were held at low density in bags (1-6), while abundant species such as *Agroiconota propinqua*, were held in high density (> 6) in each bag. Since adult density could easily affect parasitoid attraction, it may be important in future studies to keep adult density in bags as nearly constant as possible.

When retrieved from the field both the bags and vegetation within were carefully inspected for eggs. The eggs that were found on the foliage or on the mesh within these bags were transferred to Petri plates which were then placed within

plastic bags and held in the lab until larvae or parasitoids emerged or until it was determined that the eggs were not fertile or had been physically damaged.

Additionally, adult and larval Cassidinae were hand collected in the field as they were discovered and transferred to the laboratory where they were kept until parasitoids emerged or until pupation.

Once parasitoids emerged they were placed in 95° ethanol and kept at –20 °C.

Voucher specimens for all hosts and parasitoids species reared in this study have been deposited at the Smithsonian Tropical Research Institute in Panamá. Molecular seqences for the ITS2, cytochrome b and 28s genes for all Eulophidae in this study have been deposited in GenBank (see Cuignet *et al.* 2007, for accession numbers).

3.   Results

3.1.   Egg stage

Parasitoid association data were obtained for 47 Panamanian Cassidinae species, 35 of those the result of weekly sampling of sites between 2000 and 2002, while 12 others resulted from collections by DW in years dating back to 1975 in lowland central Panama (Appendix). A total of 26 parasitoid species were reared from the egg stage of Cassidinae species, 20 of the species (77%) belonged to the genus *Emersonella* (Hymenoptera, Eulophidae) and included 13 new species described by Hansson (2002).

Eulophidae (*Emersonella, Horismenus, Aprostocetus* and *Closterocerus,* in order of decreasing abundance) and Signiphoridae (one individual) were the sole Hymenopteran families reared from the egg stage of Panamanian Cassidinae. We examined 285 egg-masses from eight species of Cassidinae, out of which 21.9% (SD = 16.56) were parasitized (ie. produced one or more parasitoids (Table 1). While the exact numbers of parasitoids emerging from egg-masses was not recorded in most cases, casual observations indicated that normally more than half of eggs in parasitized egg-masses produced wasp parasitoids (MC, personal observation). Among the eight egg-mass producing species, *Chelymorpha alternans* was the most often parasitized, with 50% of its egg-masses producing one or more parasitoids. Lower percentages were observed for the maternal care species, *Acromis sparsa* (7.1%), and the nonmaternal care species, *Charidotis abrupta* 11.8%) and three *Stolas* species combined (34%). None of 31 *Chersinellina heteropunctata* egg masses were parasitized and numerous examinations of egg masses in other years and sites have yielded no evidence of an associated egg parasitoid. This monophagous species feeds on the newest, red, apical leaves of *Bonamia trichantha* Hallier f., and is the only Convolvulaceae-feeding cassidine in the study not associated with *Merremia umbellata* (L.) Hallier f. or *Ipomoea* spp.

The 23 solitary-egg depositing Cassidinae species in the mesh bag study produced a total of 5,430 eggs which after their average exposure of 3.5 days yielded 255

Species depositing:	No. eggs or egg-masses	No. parasitized	% parasitized	No. parasite spp.
Egg-masses				
Acromis sparsa	56	4	7,1	1
Charidotis abrupta	17	2	11,8	2
Chelymorpha alternans	152	76	50,0	4
Chersinellina heteropunctata	31	0	0,0	0
Cistudinella foveolata	7	1	14,3	3
Stolas lebasi	4	1	25,0	2
Stolas pictilis	15	5	33,3	1
Stolas xanthospila	3	1	33,3	1
Subtotals and averages	285	90	21.9 (sd=16.56)	
Single eggs				
Agroiconota propinqua	1526	*144*	9,4	4
Agroiconota judaica	287	*2*	0,7	1
Charidotella sexpunctata	147	*7*	4,8	2
Charidotella proxima	115	*5*	4,4	1
Charidotella sinuata	630	*23*	3,7	2
Charidotella ventricosa	138	*13*	9,4	1
Charidotella zona	125	*10*	8,0	3
Charidotis vitreata	128	*5*	3,9	1
Coptocycla leprosa	193	*0*	0,0	0
Deloyala guttata	964	*19*	2,0	4
Metrionella erratica	271	*6*	2,2	2
Microctenochira cumulata	128	*0*	0,0	2
Microctenochira flavonotata	2	*1*	50,0	0
Microctenochira fraterna	211	*9*	4,3	2
Microctenochira nigrocincta	31	*2*	6,5	2
Microctenochira sp.	144	*0*	0,0	1
Microctenochira nr nigrocincta	189	*4*	2,1	2
Microctenochira sp3	40	*0*	0,0	1
Microctenochira vivida	–	–	–	–
Spaethiella sp.	27	*1*	3,7	2
Tapinapsis atroannulus	9	*3*	33,3	1
Xenocassis ambita	117	*1*	0,9	1
Xenocassis puella	8	*0*	0,0	0
Subtotals and averages	5428	254	4.7 (sd=7.22)	

Table 1.   Number of egg-masses and eggs bagged in the field, number and percentage of eggs and egg-masses parasitized by at least one wasp after the bags were removed 7 days later, and number of cases in which phoresy was observed for each of 31 species of Panamanian tortoise beetle species.

parasitoids, an average parasitization rate of 4.7% (sd = 7.22) (Table 1). The percentage of eggs parasitized across all species from which at least 100 eggs were obtained from mesh bag exposure averaged 7.8% (N = 16, sd = 12.14%). *Agroiconota propinqua* (F.) and *Charidotella ventricosa* (Boheman) had the highest percentages of eggs parasitized, 9.4% (n = 1526) and 9.4% (n = 138), respectively. Among these 16 Cassidinae species, *Coptocycla leprosa* (Boheman) (n = 193), *Microctenochira cumulata* (Boheman) (n = 128) and *Microctenochira* sp. (n = 144) eggs produced no parasitoids. Generally, parasitoids appeared to be more efficient at finding and obtaining at least one offspring from egg-masses (22%) than they were from solitary eggs (5%). This comparison, however, may confound oviposition strategy with systematic position of the taxa involved, as six of eight 'egg-mass' species are in the tribe Stolaini, whereas all 23 'single egg' depositing species are within the more basal tribe, Cassidini.

Phoretic behavior was confirmed in 6 of the 26 egg parasitoids (*Emersonella planiceps* Hansson, *E. carballoi* Hansson, *E. rotunda* (Ashmead), *E. pubipennis* Hansson, *E. planiscuta* Hansson, and *E.* sp. nr species 4) (Table 2). Adult wasps posed quietly on the elytra of adult beetles for a considerable period of time, apparently waiting until the host oviposited for a chance to oviposit within a fresh egg. A maximum of four wasps were observed riding on the elytra of adult beetles. In cases where both phoretic and reared parasitoids were identified for a particular beetle species, the phoretic species was always within the largest set of species reared from the eggs of that beetle. Phoretic parasitoids were observed and collected from the elytra of 15 out of 47 (32%) Cassidinae species (Table 2). Two phoretic species were associated with hosts which deposit egg-masses, and four with hosts which deposit single-eggs. *Emersonella planiceps* was observed and collected from the elytra of 11 Cassidinae species and successfully reared from the eggs of five of those species. *Emersonella rotunda*, on the other hand, was collected from only one host, *Charidotella zona*, but was reared from the eggs of 13 cassidine species (Appendix).

Among the 20 species of *Emersonella* reared from egg masses of 47 species of Cassidinae, 12 species (*Emersonella albicoxa* Hansson, *E. varicolor* Hansson, *E. reticulata* Hansson, *E. horismenoides* Hansson, *E. cuignetae* Hansson, *E. desantisi* Hansson, *E. tanigaster* Hansson, *E. planiscuta*, *E.* sp. nr *hastata*, *E.* sp. 1, *E.* sp.2 *E.* sp. 5) were reared from a single host species and thus appear to be potential host specialists (Appendix). *Emersonella pubipennis* was reared from two related subsocial species of Stolaini, *Acromis sparsa* (Boheman) and *Paraselenis tersa* (Boheman). *Emersonella niveipes* parasitized a number of other species of Stolaini in the genera, *Stolas* and *Chelymorpha*. *Emersonella planiceps* was mainly associated with the genus *Microctenochira*, and *E. rotunda* with the Cassidini genera, *Charidotella*, *Charidotis* and *Agroiconota*. *Emersonella planiceps* was exclusively associated with the genus *Microctenochira* during the first two sampling periods (November to December 2000, and May to June 2001) but during the third sampling period (May and Jun. 2002) it commonly emerged from eggs of *Charidotella sinuata*, and in one

Phoretic parasitoid species	Cassidinae host species	Total number of hosts obs.	Parasitoid reared	Host oviposition*	No. of phoretic parasitoids	No. of hosts with parasitoids
Emersonella pubipennis	Acromis sparsa	209	yes	m	8	7
Emersonella planiscuta	Stolas lebasi	9	no	m	2	1
Emersonella carballoi	Agroiconota propinqua	480	yes	s	5	4
Emersonella planiceps	Agroiconota propinqua	–	yes	s	2	1
	Agroiconota judaica	135	no	s	2	1
	Charidotella sexpunctata	84	no	s	4	2
	Charidotella sinuata	83	yes	s	18	11
	Microctenochira fraterna	293	yes	s	10	7
	Microctenochira sp1	153	yes	s	13	8
	Microctenochira nigrocincta	56	yes	s	2	2
	Microctenochira nr nigrocincta	36	no	s	4	4
	Microctenochira flavonotata	14	no	s	1	1
	Microctenochira vivida	–	no	s	1	1
	Microctenochira cumulata	28	no	s	1	1
	Xenocassis ambita	92	no	s	1	1
Emersonella rotunda	Charidotella zona	48	yes	s	2	2
Emersonella nr species 4	Xenocassis ambita	–	no	s	2	1

*   m = egg-mass, s = single egg

Table 2.   List of egg parasitoid species exhibiting phoresy, their Cassidinae host species, total number of Cassidinae observed, Cassidinae species from which the phoretic parasitoids were reared from host eggs, oviposition strategy of their hosts, total number of phoretic parasitoids and number of Cassidinae upon which they were observed.

case from eggs of *Agroiconota propinqua* and *Xenocassis ambita*. Similarly, *E. rotunda*, which previously emerged exclusively from *Charidotella* and *Agroiconota* eggs, emerged occasionally during the third sampling period from *Microctenochira* species. These apparent temporal shifts in preference were not due to any obvious change in the abundances of their beetle hosts.

3.2.   Larval and pupal stages

A total of 146 individuals in 15 parasitoid species were reared from 1999 Cassidinae larvae belonging to three Stolaini species with gregarious larvae and four Cassidini species with solitary larvae (Appendix and Table 3). All seven Cassidinae species were broadly sympatric and fed on Convolvulaceae (*Merremia umbellata* and *Ipomoea* spp.). The larvae of *Agroiconota propinqua* and *A. judaica* (F.) were indistinguishable in the field and for that reason their numbers were pooled. Larvae were chiefly parasitized by Tachinidae (*Eucelatoria* spp. and *Ebenia* spp.) and Chalcididae (*Brachymeria* spp. and *Conura* spp.). However, multiple individuals of a *Horis-*

Species	No. larvae		No. parasitoids		%
Acromis sparsa	561		75		13,4%
Chelymorpha alternans	1012		27		2,7%
Stolas pictilis	197		2		1,0%
Gregarious (Stolaini)		1770		104	5,9%
Agroiconota spp.	128		26		20,3%
Deloyala guttata	31		2		6,5%
Metrionella erratica	38		3		7,9%
Charidotella sinuata	32		11		34,4%
Solitary (Cassidini)		229		42	18,3%
All taxa		1999		146	7,3%

Table 3. Summary of numbers of parasitoids reared from the larvae of seven species of Cassidinae.

menus sp. (Eulophidae) emerged from a single *Spaethiella marginata* (Champion) larva. A single *Nematomorpha* was reared from a *Stolas cuculata* (Boheman) larva and another from an *Agroiconota propinqua* larva. The overall rate of larval parasitism for the seven Cassidinae taxa was 7.3%, with the three species with gregarious immature stages suffering less from parasitoids than those with solitary stages, 5.9 ± 6.85% (n = 3) versus 18.3 ± 8.08% (n = 4), respectively.

Eggs, larvae and pupae of the lepidopteran *Schacontia* sp. (Crambidae) were reared from infected larval assemblies of *Polychalma multicava* (Fig. 1a, 1f). Adults of this moth oviposited on the exuvia of first and second instar larvae (Fig. 1b). Once *Schacontia* larvae eclosed they traveled externally about the *Polychalma* larva, chewing the tips of scoli and feeding on the hemolymph which issued from them. *Schacontia* larvae built refuges of silk most often attached to scoli and the exuvial annex of the *Polychalma* larva, but also laterally at the junction between pronotal and abdominal segments, and occasionally on the head of the larva (Fig. 1c, 1d). The development of *Polychalma* larvae appeared to be minimally affected by their parasite until the moth larvae approached pupation. At that point *Schacontia* began to use silk to ensnare adjacent larvae and they switched from feeding externally on hemolymph to feeding internally on the organs of their captured hosts. They then pupated in lightly spun cocoons to which fragments of larval head capsules, scoli and pupal fragments remained attached (Fig. 1e). Although *Schacontia* larvae seemed capable of successfully developing on a single *Polychalma* larva or pupae, the gregarious habits of its host nearly always supplied several bodies for the final feast. While *Schacontia* appeared to be nearly an exclusive parasite of *Polychalma* larvae, one larva was discovered feeding on an *Acromis* larval aggregation, opening the possibilities that additional species exist or that this particular species has greater dietary flexibility than our encounters of it on *Polychalma* would suggest.
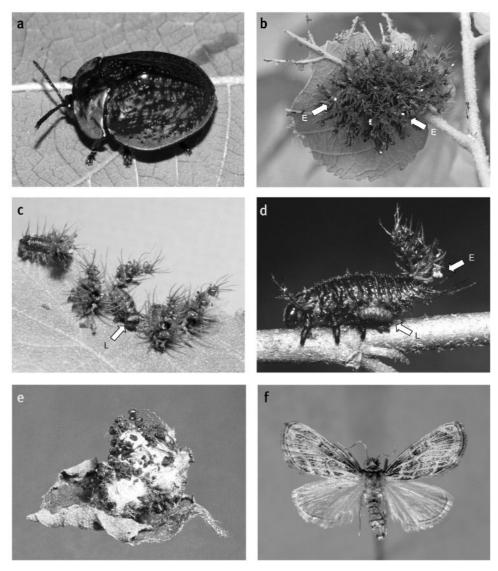
Figure 1. *a.* adult *Polychalma multicava*; *b.* a typical group of second instar *P. multicava* larvae on *Helicteres guazumifolia* (Sterculiaceae) bearing the eggs (E) of *Schacontia* sp. (Lepidoptera, Crambidae); *c.* a loose association of fourth instar *P. multicava* larvae, one of which bears the larva (L) of its koinobont parasitoid; *d.* a fifth instar *P. multicava* larva with a *Schacontia* larva (L) in a lateral retreat; *e. Schacontia* sp. puparium composed of silk and adhering sclerites of two or more *Polychalma* larvae; *f.* adult *Schacontia* sp. specimen reared from *Polychalma* larvae.

3.3.   Adult stage

Tachinidae appear to parasitize the larval stages of smaller Cassidinae species, especially those in the tribe Cassidini (*Agroiconota, Charidotella, Microctenochira*, etc.). However, some *Eucelatoria* species also are important parasitoids of the adult stage

Collecting site	Province	Collection dates	Chely-morpha alternans	Tachi-nidae	Nemato morpha	Other	% Tachi-nidae
I. Censuses:							
Gam/Camp/Met	Panama	2xi-24xii00; 2v-30vi01; 3v-2vii02	289	12	0	–	4,2%
II. Lab reared adults:							
Cerro Campana	Panama	16ix,5x,9xi02; 11v,14xii03	92	1	0	21	1,1%
Cana/Patino	Darien	11xii04, 9vii05	37	3	0	10	8,1%
Chiriqui Grande	Bocas del Toro	8vii05	70	5	0	3	7,1%
Guarumal	Cocle	31vii03; 1vii,10x04	82	7	0	14	8,5%
Las Lajas	Chiriqui	1ix,9xi03; 10x04	93	0	3	14	0,0%
Remedios	Chiriqui	1v,29vi,9xi03; 1vii,10x04	270	24	3	26	8,9%
Gamboa/Pina	Colon	8ix,25x02; 27vii,10viii,24viii04	79	0	0	1	0,0%
Subtotal			723	40	6	89	4.8% avg. (n=7)
III. Dissected adults:							
Remedios	Chiriqui	1vi04	50	16	2	–	32,0%
Gamboa	Colon	29vii04…12viii04	29	1	0	–	3,4%
Cerro Campana	Panama	4vii04…25vii04	32	16	0	–	50,0%
P. Metropolitano	Panama	24vii04…21viii04	12	3	0	–	25,0%
Subtotal			125	36	2	–	27.6% avg. (n=4)

Table 4. Occurrence of Tachinidae (Diptera), *Nematomorpha* and other parasites in collections of *Chelymorpha alternans* adults. The first census results refer to beetles collected during the periods of the mesh bag studies and maintained for short period in lab conditions, the second to beetles collected in 6 provinces of Panama and maintained in lab culture for 1-2 months, the third to beetles collected, maintained for 1-2 weeks in the lab and then dissected. In the first two censuses only Tachinidae emerging and pupating were scored, while in the third census all beetles were examined for the presence of larval Tachinidae in the abdomen.

in at least one Stolaini, *Chelymorpha alternans*. Tachinid larvae emerged from 12 of 289 (4.2%) *C. alternans* adults collected as larvae on weekly censuses during 2000 and provisioned with surface-sterilized *Merremia* leaves (Appendix). The beetles were alive when the tachinid larva emerged through slits in the intersegmental membranes of the adomen but died usually within minutes or a few hours as most abdominal organs were consumed. Additional censuses of *C. alternans* populations were performed across a wider array of sites in 2002 and 2004 with beetles maintained for a minimum of 30 dy following collection. Forty Tachinidae emerged from 723 adult beetles, an infection rate of 4.82%, a rate remarkably similar to that obtained by the first censuses (Table 4-11). However, because considerable adult mortality was not explained by tachinid emergence, 125 additional *C. alternans*

adults were collected at 4 sites during June through August 2004 (Table 4). More than 27% of these beetles produced either a tachinid larva while being held in the lab or tachinid larvae were obvious in the abdomen upon dissection. Ovaries were present and developed in 75% of non-parasitized female beetles but were absent in females containing tachinid larvae. Testes were entirely missing in 10 of 11 males containing tachinid larvae. While multiple tachinid larvae were often observed in the abdominal cavity of the host, only a single larva was ever observed to emerge from an adult beetle or pupa suggesting that larval canibalism must be occurring fairly commonly within the abdomens of the beetles.

Among Tachinidae emerging from larval and pupal stage Cassidinae, *Eucelatoria* sp1 was associated with *Discomorpha salvini* (Baly), *Omocerus casta* (Boheman) and *Physonota alutacea* Boheman. *Eucelatoria* sp3 parasitized the Stolaini species, *Omaspides bistriata* and *Stolas xanthospila* Champion. *Eucelatoria* sp2 emerged from larval *Cistudinella foveolata* Champion and *Paraselenis tersa*, which belong to different tribes and feed on host-plants of different families. Three additional unidentified Tachinidae species were reared from larval *Chelymorpha alternans*.

Two unidentified Nematomorpha were reared separately from an adult *Stolas cuculata* and from a female *Acromis sparsa* (out of 209 specimens collected) (Table 1). Eight additional *Nematomorpha* were obtained from *Chelymorpha alternans* adults collected on the 2002-2005 censuses. The relatively small numbers of *Nematomorpha* encountered in all three censuses suggests they are a less serious mortality factor than Tachinidae.

4.   Discussion

For many leaf beetles, the egg stage may be poorly defended and vulnerable to predators and parasitoids (Selman 1994). Insect eggs generally lack self-generated secondary compounds or secondary compounds derived from plants (Gauld & Gaston 1992; Pasteels *et al.* 1988; Hilker 1994; Eben 1999). Further, eggs may lack a cellular defense response to foreign intrusion (Strand 1986). Cassidinae eggs are heavily parasitized in the Paleotropics by Encyrtidae (Simon Thomas 1964), Eulophidae (*Tetrastichus* spp.) and Tetracampidae (*Cassidocida* spp.) (Nakamura & Abbas 1989), and possibly other taxa when wider surveys are undertaken. Based on the reports of Becker & Frieiro Costa (1987, 1988) and Carrol (1978), the most common egg parasitoids of Cassidinae in the Neotropics are species of *Emersonella* (Eulophidae). Indeed, our study documents that Eulophidae and one Signiphoridae attack the egg stage of at least 35 out of 41 Panamanian Cassidinae. Twenty species of *Emersonella* (Eulophidae) were reared from the eggs and oothecae of 33 out of 41 tortoise beetles species. Our results further indicate that *Emersonella* are abundant, have a great impact on egg survival and may well have had a long history of diversification on their tortoise beetle hosts (Cuignet *et al.* 2007).

Olfactometer tests indicate that Eulophidae are not attracted by cues coming from the eggs of the host, but that encounter is largely by chance (Hilker & Meiners

1996; Meiners & Hilker 1997; Meiners *et al.* 2000; and personal observations). How, then, do Eulophidae locate the eggs that they parasitize? More than sixty percent of the Cassidinae species in our study were observed bearing phoretic parasitoids at least once during this study. Six of the 20 *Emersonella* species collected in our mesh-bag study were phoretic, and phoretic wasps were observed on 15 out of the 41 Cassidinae in the study, indicating the prevalence of phoresy. Previously, phoresy was reported in only two species of Eulophidae (Gauthier *et al.* 2000). One of these species, *E. niveipes*, has been reported on several *Stolas* sp. (Carroll 1978; Hilker & Meiners 1996) but was not observed to be phoretic on *Chelymorpha alternans* and *Stolas pictilis* in this study.

Clausen (1976) hypothesized that species known to be phoretic should attack host species that deposit their eggs in masses. Four of the six phoretic parasitoids in our study were associated with Cassidinae species that deposited eggs singly indicating that this is not exclusively the case in Cassidinae. The low percentage of parasitized eggs we recorded for species laying solitary eggs suggests that, although it may be cost-effective for parasitoids to adopt a phoretic strategy to shorten host discovery time, higher rates of parasitism are not always produced. Determining whether phoresy is a flexible strategy, employed when host densities are low and abandoned when hosts are common could be an interesting direction for future studies.

Published studies do indicate the egg stage of Cassidinae is heavily parasitized (Becker & Frieiro Costa 1988; Carroll 1978; Windsor 1987; Obermaier *et al.* 2000; Nakamura & Abbas 1987, 1989). Whether egg-masses are more vulnerable to egg parasitoids than solitary eggs is difficult to evaluate because there are few comparable studies conducted at the same place and time for both single-egg and multiple-egg depositing species. The parasitism rates we obtained for single-egg depositing species (for which there were 10 or more observations) ranged from 0 to 9.4% (Mn = 3.5%) and from 0 to 50% (Mn = 21.9%) for egg-mass depositing species. However, there is little indication that the number of parasitoid species attacking solitary and egg-mass depositing species differs significantly (ie. ranges from 0 to 4 in both groups; Table 1). The lower rates of egg discovery in single-egg depositing species may be due to a number of factors, including, 1) single eggs are less conspicuous to visually hunting parasitoids, 2) fewer volatiles or synomones are released by single eggs during and immediately following oviposition, 3) single-egg depositing beetle species tend to be smaller and give off fewer odour cues for parasitoids and move more quickly over the food plant and thereby leave a less concentrated and more complex set of cues for parasitoids.

Parental care may act to reduce egg-mass parasitism and, indeed, in this study the egg masses of the subsocial Stolaini species, *Acromis sparsa*, were less parasitized than egg masses of the non-subsocial *Chelymorpha alternans* and *Stolas pictilis* (Table 1). However, *Acromis sparsa* studied earlier by Windsor (1987) was parasitized at a rate ten times higher than reported here (77% vs 7%). This large discrepancy may represent a difference in protocol between the two studies, with

the earlier study calculating losses to parasitoids after the entire egg development period, and the later study determining losses from egg-masses moved into the lab well before larval ecolosion. Time of year may also influence the efficiency of parasitoids as their populations appear to increase over the course of the wet season. The earlier study was conducted in the late wet season (August through November), while egg masses examined in the present study came primarily from the early wet season (May-July).

The Eulophidae species attacking the egg masses of subsocial Stolaini species (*Acromis sparsa*, *Paraselenis tersa*, *Omaspides bistriata* and *O. convexicollis*) were not found to emerge from other Cassidinae. Indeed, *A. sparsa* and *P. tersa* share the same parasitoid (*Emersonella pubipennis*) and the two *Omaspides* species also share a single parasitoid (*E. windsori*). These two eulophid species fall within the same clade in a Matrix Representation Parsimony (MRP) super-tree, with the *Omaspides* wasp basal to the species attacking *Acromis* and *Paraselenis* eggs (Cuignet *et al.* 2007). We suspect that the two *Emersonella* species attacking parental care cassidines are specialists on those species, perhaps having evolved offensive tactics that enable them to circumvent the defensive behaviour of the guarding mother. Further, subsocial Stolaini appear to be monophagous while other Cassidinae are narrowly oligophagous. This dietary specialization in maternal care species also could favour wasp sensory specialization making host escape more difficult than in oligophagic species. Thus, parental care and other host behaviors could influence the guild composition of parasitoids by favouring specialists species and eliminating generalists from the list of species attacking particular host beetle species. Additional collecting and identification of Eulophidae from parental-care and non-parental-care tortoise beetle species will be crucial to testing these possibilities.

Larvae of seven Cassidinae species taken from the field and reared in the lab were parasitized chiefly by Chalcididae (*Brachymeria* spp. and *Conura* spp.) and by Tachinidae (*Eucelatoria* spp., *Ebenia* spp. and unidentified Tachinidae species) (Appendix). The gregarious larvae of *Chelymorpha alternans* and *Stolas pictilis* were less frequently parasitized by both Chalcididae and Tachinidae than the maternally-protected larvae of *Acromis sparsa*. Predators, especially the pentatomid bug, *Stiretrus anchorago* (Asopinae), were a large but unmeasured source of larval mortality for many species in this study. Future studies that expose larvae to both the parasitoid and predator communities while controlling the exposure time, age of larvae and period of the year could be highly informative of the complex physical and biotic factors affecting Cassidinae survival.

As their name suggests, tortoise beetles can retract all appendages beneath the elytra and pronotum when disturbed (Olmstead 1996). Tarsal claws and pads composed of minute secretory setae allow them to pull the carapace flush with the substrate (Eisner 1971; Windsor *et al.* 1992). The pronotum and elytra are heavily sclerotized and the ovipositor of parasitoids such as Tachinidae and the mouthparts of predatory Heteroptera may be unable to penetrate to the soft parts. Thus, the main portal for parasites attacking the adult stage may be through larval ingestion

of tachinid eggs or by oviposition by Tachinidae directly onto or into larvae (Belshaw 1994). That Tachinidae are infecting larvae is evidenced by numerous cases in which we brought larvae into the lab, fed them surface-sterilized leaves of their foodplant, observed them pupate and eclose as adults and within a few weeks die from an exiting tachinid larva. The rapidity with which some infected adult beetles die after their eclosion suggests the tachinid larva is already present, perhaps as an egg, within the adult when it ecloses from pupation. The finding that there is no significant difference in the length of parasitized and non-parasitized beetles additionally suggests that tachinids do not immediately begin to prey on the immature stages of the beetles they have infected (Reardon 2005). Instead, some Eucelatoria species larvae appear to target the developing gonads, soft tissues and fat deposits in the abdomens of young adult *Chelymorpha alternans*. More than 25% of adult, field-collected *C. alternans* were parasitized by Tachinidae.

As suggested by Price (1980) the potential for specialization among parasites is high when the environment is discontinuous, when host diversity is high and when resources are transient. A considerable amount of specialization may be evident in the eulophid parasitoid community that we sampled. We reared 11 of 15 Eulophidae species from the eggs of single hosts. While we acknowledge that some of these records could be the product of small sample size or low parasite population levels, some degree of specialization is indicated by the multiple independent rearings of *Emersonella cuignetae* on *Chelymorpha alternans* and *E. albicoxa* on *Hilarocassis* and the absence of these parasitoids in the rearings of other species. Additionally, these apparently host-specific parasitoids are associated more often than one might expect with egg-mass producing host species (11 out of 14 parasitoid species). While the majority of Eulophidae appear to be specialists, some species are clearly oligophagous. *E. planiceps* and *E. rotunda* were reared from 11 and 13 different hosts respectively. Nevertheless, they emerged exclusively from single-egg depositing beetle species, in the *Microctenochira* and *Charidotella* species groups. So, while these two parasitoid species accept large numbers of host species, their hosts are closely related and not a random selection of Cassidinae available in the habitat. However, larger samples of species and tests which minimize autocorrelation due to common ancestry will be needed before definitive patterns can be entirely accepted.

5. Conclusion

Despite the numerous and diverse defense mechanisms deployed by Neotropical Cassidinae, our survey of 47 species reveals they are indeed heavily parasitized and that claims may not be wrong that they are the most heavily parasitized chrysomelid subfamily, however, it will be difficult to objectively evaluate this claim. A diverse group of Eulophidae species including host specialists and generalists attacked the egg stage of 35 of 47 species. Chalcidid wasps (*Brachymeria* and *Conura* species), tachinid flies (diversity largely unstudied), *Nematomorpha* worms and one ectoparasitic lepidopteran (Crambidae) attacked larval stages of various

species. Larval Tachinidae were dissected from the abdomens of more than a fourth of all adult *Chelymorpha alternans* examined. A large proportion of the parasitoid species we collected were undescribed, indicating that a wealth of parasitoid diversity probably still remains to be discovered and documented in other tropical Chrysomelidae. Even within the species we have already studied, molecular sequencing is needed to better understand the patterns of diversity existing in hosts and parasitoids.

Acknowledgements

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Appendix

Host-parasitoid associations. Numbers inside the table correspond to the total number of parasitoids collected with the number of independent collections performed indicated by parentheses. DW = collected by D. Windsor in preceding years. Host-plant family: Con = Convolvulaceae, Bor = Boraginaceae, Ast = Asteraceae, Hel = Heliconiaceae. Family of the parasitoid: S = Signiphoridae, Nemato = Nematomorpha, Cram = Crambidae (Lepidoptera). Stage attacked: E = Egg, L = Larva, L-P = Larva and pupa, L-A = Larva to adult, P = Pupa, A = Adult. Ph = Phoresy observed. Darkened names indicate beetle species that deposit egg-masses, the other taxa disperse eggs singly.

Parasitoid family: …Eulophidae…

Cassidinae host species	Host abundance	HP family	E. albicoxa (new sp.)	E. carballoi (new sp.)	E. cuignetae (new sp.)	E. desantisi	E. horsmenoides (new sp.)	E. nivetipes	E. nr hastata (new sp.)	E. planiceps (new sp.)	E. planiscuta	E. pubipennis (new sp.)	E. rotunda	E. reticulata	E. tangaster	E. varicolor	E. windsori (new sp.)	E. sp. 1 (new sp.)	E. sp. 2 (new sp.)	E. sp. 3 (new sp.)	E. sp. 4 (new sp.)	E. sp. 5 (nr carballoi) (new sp.)	Horismenus sp. 1	Horismenus sp. 2
Stage attacked			E Ph	E Ph	E	E	E	E	E	E Ph	E Ph	E Ph	E Ph	E	E	E	E	E	E	E	E Ph	E	E	E
1 Acromis sparsa	209	Con	.	.	.	.	.	.	.	5(4)	.	52(8)	.	.	.	.	.	.	.	.	.	.	.	.
2 Agroiconota propinqua	480	Con	.	3(2)	.	.	.	.	.	.	.	.	139(29)	.	.	.	.	.	.	.	.	.	.	.
3 Agroiconota judaica	135	Con	.	.	.	.	.	.	.	.	.	.	4(3)	.	.	.	.	.	.	.	.	.	.	.
4 Charidotella sexpunctata	84	Con	.	.	.	.	.	.	.	.	.	.	7(3)	.	.	.	.	.	.	.	.	.	.	.
5 Charidotella proxima	118	Con	.	.	.	.	.	.	.	4(2)	.	.	3(3)	.	.	.	.	.	.	.	.	.	.	.
6 Charidotella sinuata	83	Con	.	.	.	.	.	.	.	24(15)	.	.	14(6)	.	.	.	.	.	.	.	.	.	.	.
7 Charidotella ventricosa	18	Con	.	.	.	.	.	.	.	.	.	.	12(2)	.	.	.	.	.	.	.	.	.	.	.
8 Charidotella zona	48	Con	.	1(1)	.	.	.	.	.	.	.	.	7(6)	.	.	.	.	.	.	.	.	.	.	.
9 Charidotella sp.	1	Con	.	1(1)	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
10 Charidotis abrupta	2	Bor	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
11 Charidotis vitreata	16	Bor	.	.	.	2(2)	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
12 Chelymorpha alternans	289	Con	.	.	18(3)	.	.	456(25)	.	.	.	.	4(1)	.	87(3)	.	.	17(2)	.	.	.	.	.	.
13 Chelymorpha sericea	DW	Con	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
14 Chelymorpha gressoria	9	Con	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
15 Chersinellina heteropunctata	8	Con	.	.	.	.	15(3)	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
16 Cistudinella foveolata	5	Bor	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	8(1)	.	.
17 Coptocycla leprosa	4	Bor	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
18 Deloyala guttata	174	Con	.	5(3)	.	.	.	.	.	.	.	.	3(2)	.	.	.	.	.	.	11(5)	.	.	.	.
19 Discomorpha salvini	DW	Bor	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	1(1)	.	.	.
20 Eugenysa coscaroni	DW	Ast	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
21 Hilarocassis evanida	DW	Con	18(3)	.	.	.	.	18(1)	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
22 Hybosa melicula	DW	Big	.	.	.	.	.	.	7(1)	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
23 Iscnocodia annulus	13	Bor	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
24 Metrionella erratica	57	Con	.	.	.	.	.	.	.	.	.	.	3(1)	.	.	.	.	.	.	1(1)	.	.	.	.
25 Microctenochira cumulata	28	Con	.	.	.	.	.	.	.	1(1)	.	.	.	.	.	.	.	.	.	.	.	.	.	.

Parasitoid family: ...Eulophidae...

Cassidinae host species	Host abun-dance	HP fam-ily	*Emersonella albitoxa* (new species)	*Emersonella carballoi* (new species)	*Emersonella cuignetae* (new species)	*Emersonella desantisi*	*Emersonella horismenoides* (new species)	*Emersonella niveripes*	*Emersonella* nr *hastata* (new species)	*Emersonella planiceps* (new species)	*Emersonella planiscuta* (new species)	*Emersonella pubipennis* (new species)	*Emersonella rotunda*	*Emersonella reticulata*	*Emersonella tanygaster*	*Emersonella varicolor*	*Emersonella windsori* (new species)	*Emersonella* sp. 1 (new species)	*Emersonella* sp. 2 (new species)	*Emersonella* sp 3 (new species)	*Emersonella* sp. 4 (new species)	*Emersonella* sp. 5 (nr carballoi) (new species)	*Horismenus* sp. 1	*Horismenus* sp. 2
Stage attacked			E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E	E
HP family			Ph	Ph						Ph	Ph	Ph	Ph								Ph			
26 *Microctenochira flavonotata*	14	Ast																						
27 *Microctenochira fraterna*	233	Con								5(5)			7(3)											
28 *Microctenochira infantula*	9	Con								1(1)														
29 *Microctenochira* sp1	153	Con								14(8)														
30 *Microctenochira* sp3	9	Con								2(2)														
31 *Microctenochira nigrocincta*	56	Con								3(3)			1(1)											
32 *Microctenochira* nr *nigrocincta*	36	Con											6(4)											
33 *Omaspides bistriata*	DW	Con															3(1)							
34 *Omaspides convexicollis*	DW	Con															4(1)							
35 *Omocera casta*	DW	Bor																						
36 *Paraseleis tersa*	DW	Con																						
37 *Physonota alutacea*	DW	Bor										2(1)												
38 *Polychalma multicava*	DW	Bor												20(1)								8(1)	8(1)	8(1)
39 *Spaethiella marginata*	116	Hel																						
40 *Stolas cuculata*	9	Con																						
41 *Stolas lebasi*	9	Ast						77(8)			2(1)							30(1)						
42 *Stolas pictilis*	40	Con						8(2)																
43 *Stolas xanthospila*	8	Con														9(1)								
44 *Stolas* sp.	DW	Con																						
45 *Tapinaspis wnesmali*	3	Ast																			2(1)			
46 *Xenocassis ambita*	92	Con								1(1)														
47 *Xenocassis puella*	52	Con																						
Number of host species:	1		4	1	1	1	4	1	11	1	2	13	1	1	1	2	1	1	2	2	1	1	1	3
Number of parasitoids:			18	10	18	2	15	159	7	62	2	54	186	20	87	9	7	17	30	12	3	8	8	8

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#	Cassidinae host species	Host abun-dance	HP family	Horismenus sp. 3	Aprostocerus sp.	Closterocerus sp.	Signiphoridae Unid.	Brachymeria russelli	Brachymeria sp. (grp russelli, nsp.)	Conura sp. N°374 (grp immaculata)	Conura sp (new group)	Chalcididae Unid.	Euclatorria nana (?)	Euclatorria sp. 1	Euclatorria sp. 2	Euclatorria sp. 3	Eberia sp. 1	Eberia sp. 2	Tachinidae Unid. 1	Tachinidae Unid. 2	Nematophora sp. 1	Nematophora sp. 2	Schaconia sp. (Lep., Crambidae)
Parasitoid family:				S		...Chalcidae...									...Tachinidae...						Nemato		Cram
Stage attacked				L	E	E	E	L-P	L-P	L-P	L-P	L-P	L	L	L	L	L	L	L-A	L-A	A	L	L
1	Acromis sparsa	209	Con	·	·	·	·	·	·	·	·	·	·	34(4)	·	·	·	·	·	·	·	·	·
2	Agroiconota propinqua	480	Con	·	1(1)	·	·	·	·	·	·	·	·	·	·	·	·	·	26(10)	1(1)	1(1)	1(1)	·
3	Agroiconota judaica	135	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
4	Charidotella sexpunctata	84	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
5	Charidotella proxima	118	Con	·	·	·	·	·	·	·	·	1(1)	·	·	·	·	·	·	10(6)	·	·	·	·
6	Charidotella sinuata	83	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	1(1)	·	·	·	·
7	Charidotella ventricosa	18	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
8	Charidotella zona	48	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
9	Charidotella sp.	1	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
10	Charidotis abrupta	2	Bor	·	·	·	9(1)	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
11	Charidotis vitreata	16	Bor	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
12	Chelymorpha alternans	289	Con	·	·	·	·	15(5)	·	·	·	·	·	·	·	·	2(2)	·	9(7)	1(1)	·	·	·
13	Chelymorpha sericea	DW	Con	·	·	·	·	1(1)	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
14	Chelymorpha gressoria	9	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
15	Chersinellina heteropunctata	8	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
16	Cistudinella foveolata	5	Bor	2(1)	·	·	·	·	·	·	·	·	·	·	1(1)	·	·	·	·	·	·	·	·
17	Coptocycla leprosa	4	Bor	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
18	Deloyala guttata	174	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	2(2)	·	·	·	·
19	Discomorpha salvini	DW	Bor	5(1)	1(1)	·	·	·	·	·	·	·	·	1(1)	·	·	·	·	·	·	·	·	·
20	Eugenysa coscaroni	DW	Ast	·	·	·	·	·	1(1)	·	·	·	·	·	·	·	·	·	·	·	·	·	·
21	Hilarocassis evanida	DW	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
22	Hybosa melicula	DW	Big	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
23	Iscnocodia annulus	13	Bor	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
24	Metrionella erratica	57	Con	·	·	·	·	·	·	·	·	3(1)	·	·	·	·	·	·	·	·	·	·	·
25	Microctenochira cumulata	28	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·

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Cassidinae host species	Host abun-dance	HP family	Horismenus sp. 3	Aprostocetus sp.	Closterocerus sp.	Signiphoridae Unid.	Brachymeria russelli	Brachymeria sp. (grp russelli, nsp.)	Conura sp. N°374 (grp immaculata)	Conura sp (new group)	Chalcididae Unid.	Euselatoria nana (?)	Euselatoria sp. 1	Euselatoria sp. 2	Euselatoria sp. 3	Ebenia sp. 1	Ebenia sp. 2	Tachinidae Unid. 1	Tachinidae Unid. 2	Nematophora sp. 1	Nematophora sp. 2	Schaconita sp. (Lep., Crambidae)
Parasitoid family:			S				...Chalcidae...					...Tachinidae...								Nemato		Cram
Stage attacked			L	E	E	E	L-P	L-P	L-P	L-P	L-P	L	L	L	L	L	L	L-A	L-A	A	L	L
26 Microctenochira flavonotata	14	Ast	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
27 Microctenochira fraterna	233	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
28 Microctenochira infantula	9	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
29 Microctenochira sp1	153	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
30 Microctenochira sp3	9	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
31 Microctenochira nigrocincta	56	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
32 Microctenochira nr nigrocincta	36	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
33 Omaspides bistriata	DW	Con	·	·	·	·	·	·	·	1(1)	·	·	·	·	·	·	·	·	·	·	·	·
34 Omaspides convexicollis	DW	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
35 Omocera casta	DW	Bor	·	·	1(1)	·	·	·	·	·	·	·	1(1)	·	1(1)	·	·	·	·	·	·	·
36 Paraseleis tersa	DW	Con	·	·	·	·	·	·	·	·	·	·	·	1(1)	·	·	·	·	·	·	·	·
37 Physonota alutacea	DW	Bor	·	·	·	·	·	·	·	·	·	·	2(1)	·	·	·	·	·	·	·	·	·
38 Polychalma multicava	DW	Bor	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	many
39 Spaethiella marginata	116	Hel	4(1)	·	·	·	·	·	·	·	·	·	·	·	·	·	1(1)	·	·	·	·	·
40 Stolas cuculata	9	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
41 Stolas lebasi	9	Ast	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
42 Stolas pictilis	40	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
43 Stolas xanthospila	8	Con	·	·	·	·	·	·	1(1)	·	·	·	·	·	1(1)	·	·	1(1)	·	·	·	·
44 Stolas sp.	DW	Con	·	·	·	·	·	·	·	·	1(1)	·	·	·	·	·	·	·	·	·	·	·
45 Tapinaspis waesmali	3	Ast	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	1(1)	1(1)	·
46 Xenocassis ambita	92	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
47 Xenocassis puella	52	Con	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·	·
Number of host species:	1		2	1	1	2	1	2	1	2	1	4	2	2	1	1	6	1	2	2	2	32
Number of parasitoids:			11	2	1	9	16	1	2	1	4	1	38	2	2	2	1	49	1	2	2	many	124