Native parasitoids associated with *Tuta absoluta* in the tomato production areas of the Spanish Mediterranean Coast

Rosa Gabarra · Judit Arnó · Lidia Lara · María Jesús Verdú · Antoni Ribes · Francisco Beitia · Alberto Urbaneja · María del Mar Téllez · Oscar Mollá · Jordi Riudavets

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Abstract The tomato borer *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is an invasive pest that produces significant damage to tomato crops in the Mediterranean area. Although several species of predatory bugs are successfully being used for biological control of the pest, little is known about the parasitoids that are able to exploit *T. absoluta* as a host. With the aim of better understanding parasitoid species richness of *T. absoluta* along the Mediterranean Spanish Coast, we conducted an extensive survey to determine distribution, host plants and habitats where parasitoids are present. Our results indicated

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R. Gabarra · J. Arnó (⊠) · J. Riudavets Entomology, Institut de Recerca i Tecnologia Agroalimentària (IRTA), Ctra. de Cabrils, km 2, 08348 Cabrils, Barcelona, Spain e-mail: judit.arno@irta.cat

L. Lara · M. d. M. Téllez Instituto de Investigación y Formación Agraria y Pesquera (IFAPA), Camino de San Nicolás, n°1, 04745 La Mojonera, Almería, Spain

M. J. Verdú · F. Beitia · A. Urbaneja · O. Mollá Departamento de Entomología, Centro de Protección Vegetal y Biotecnología, Instituto Valenciano de Investigaciones Agrarias (IVIA), Ctra. Moncada-Náquera, km 4.5, 46113 Moncada, Valencia, Spain

A. Ribes

C. Lleida, 36. Torres de Segre, 25170 Lleida, Spain

that egg parasitoids are naturally scarce but that the species richness of larval/pupal parasitoids is high and includes 20 different species. Seven of these had not been previously reported as *T. absoluta* parasitoids. The most frequent parasitoid species recovered were *Necremnus* sp. nr. *artynes* (Walker), *Stenomesius* cf. *japonicus* (Ashmead) and *Neochrysocharis formosa* (Westwood) (Hymenoptera: Eulophidae).

Keywords Necremnus artynes · Stenomesius japonicus · Bracon nigricans · Conservation biological control · Parasitoid richness · Noncrop habitats

Introduction

The South American tomato pinworm, or tomato borer, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is an important pest native to South America that produces significant damage to tomato crops, both in the greenhouse and open fields, throughout the growing season. Larvae of *T. absoluta* mainly attack leaves and fruits but can also feed on stems, buds and flowers, causing severe crop losses of up to 100 % when no control measures are present. In Spain, *T. absoluta* was first detected at the end of 2006 in the north of Castellón (eastern Spain) this being the first record of this species out of its continent of origin (Urbaneja et al. 2007). Since then, this invasive pest has spread rapidly throughout multiple locations in the main Mediterranean coastal tomato-producing areas, but also to some other countries in Europe, Africa and Asia (Desneux et al. 2011). In all newly infested areas, *T. absoluta* immediately reached damaging levels, despite being intensively treated with pesticides (Desneux et al. 2010). However, this broad use of pesticides initially disrupted the existing integrated pest management programs used for tomato crops (Urbaneja et al. 2012). Therefore, efforts were directed to identifying sustainable methods for biological control.

Arthropod natural enemies that inhabit a region prior to a pest invasion may establish new associations with the invasive pest and contribute to its natural control (fortuitous biological control) (DeBach and Rosen 1991). There have been some recent successes in biological control programs for invasive pests based on the use and/or conservation of native Mediterranean parasitoids and predators in vegetable crops, such as the leafminer Liriomyza trifolii (Burgess) (Diptera: Agromyzidae) and the whitefly Bemisia tabaci (Gennadius) (Hemiptera: Aleyrodidae). Liriomyza trifolii invaded the Mediterranean Basin in the early 1980s, and the native parasitoids Diglyphus isaea (Walker) and Neochrysocharis formosa (Westwood) (Hymenoptera: Eulophidae) were, and remains, crucial to its control (Gabarra and Besri 1999; Nicoli 1997). Similarly, when B. tabaci invaded vegetable crops in Spain in the 1990s, the native parasitoid Eretmocerus mundus Mercet (Hymenoptera: Aphelinidae) and the polyphagous predators Macrolophus pygmaeus (Rambur), Dicyphus tamaninii Wagner and Nesidiocoris tenuis (Reuter) (Hemiptera: Miridae) were shown to be important control agents of the pest (Arnó et al. 2010a; Gerling et al. 2001; Stansly et al. 2004). These natural enemies are currently being used in biological control programs despite their phytophagy may result in plant damage when no prey is available (Arnó et al. 2010b; Castañé et al. 2011).

Generalist predators usually take less time to become adapted to exotic pests than parasitoids (Ehler 1998), and this might have been the case for *T. absoluta*. Several species of Miridae were detected preying upon this lepidopteran immediately after its detection in several Mediterranean countries (Urbaneja et al. 2012). *Nesidiocoris tenuis* and *M. pygmaeus* have proved to be very effective as predators of *T. absoluta* eggs (Arnó et al. 2009; Urbaneja et al. 2009). Indeed, currently successful biological control programs based on the augmentation and/or conservation of these two predatory mirid bugs have made it possible to manage not only *T. absoluta* populations but also other important tomato pests, such as whiteflies (Arnó et al. 2009; Calvo et al. 2012). Nevertheless, high populations of *N. tenuis* may cause severe damage to tomato crops when prey density is low (Castañé et al. 2011). One further aspect to take into account is that these predatory bugs take over a month to get established in the crop. During this period, the use of microbials or parasitoids may allow a more immediate pest control (Cabello et al. 2012; Chailleux et al. 2013; Mollá et al. 2011).

In the Mediterranean basin to date, some native parasitoid species have been found parasitizing the eggs and larvae/pupae of T. absoluta but not the adults. Different Trichogramma spp. (Hymenoptera: Trichogrammatidae) have been recorded parasitizing T. absoluta eggs in north-eastern Spain and Sicily (Urbaneja et al. 2012; Zappalà et al. 2012). In addition, Trichogramma achaeae Nagaraja and Nagarkatti was identified in the Canary Islands as a parasitoid of Chrysodeixis chalcites (Esper) (Lepidoptera: Noctuidae) eggs and showed great potential for reducing T. absoluta populations after its commercial release (Chailleux et al. 2012; Desneux et al. 2010; Polaszek et al. 2012), although its use is not common. Furthermore, several species of larval/pupal parasitoids have also been reported and, according to the review of Urbaneja et al. (2012) and the findings of Al-Jboory et al. (2012), Doğanlar and Yiğit (2011), Ferracini et al. (2012), Loni et al. (2011) and Zappalà et al. (2012), most of them are Chalcidoids of the Eulophidae and Braconidae families. Of these, only seven species have been found on the Spanish Mediterranean coast (Urbaneja et al. 2012). Hopefully, the species richness of indigenous natural enemies that use T. absoluta will gradually increase and contribute to the regulation of pest populations, complementing the action of the predators M. pygmaeus and N. tenuis. Our present study was aimed at conducting an extensive survey in the main tomato production areas of Spain and for a broad period of time (1) to identify the parasitoid species that use T. absoluta as a host, (2) to determine in which host plants and habitats these parasitoids are present, and (3) to estimate abundance of parasitism in crop and non crop habitats.

Materials and methods

Sampling sites

The survey was conducted in three tomato production areas along the Mediterranean coast of Spain: in the north (NE) between the coordinates $42.04^{\circ}N$ $3.18^{\circ}E$, $40.67^{\circ}N$ $0.58^{\circ}E$ and $41.63^{\circ}N$ $0.60^{\circ}E$; in the central area (CE) between $40.30^{\circ}N$ $0.27^{\circ}E$, $39.39^{\circ}N$ $0.52^{\circ}W$ and $38.19^{\circ}N$ $0.43^{\circ}W$; and in the south (SE) between $36.49^{\circ}N$ $2.53^{\circ}W$, $36.42^{\circ}N$ $2.39^{\circ}W$, $36.46^{\circ}N$ $2.13^{\circ}W$ and $36.58^{\circ}N$ $2.13^{\circ}W$, by sampling naturally occurring *T. absoluta*-infested plants, and by placing sentinel plants previously infested with eggs or larvae of *T. absoluta*.

Naturally infested plants

To determine the number of parasitoid species attacking *T. absoluta*, their distribution under natural conditions and the proportion of samples with parasitism, naturally infested plants were sampled from 2008 to 2011 in the three tomato production areas mentioned above (Table 1). The survey was started in 2008 in the NE and CE, the two areas that were first invaded by *T. absoluta*, and at the end of 2009 in the SE. In the NE, most of the samples came from crops and only few were *Solanum nigrum* L. infested plants. Sampled crops were

Table 1 Number of samples collected from 2008 to 2011 in naturally infested plants in three areas located in the north (NE), central (CE) and south (SE) part of the Mediterranean coast of Spain (see coordinates in the text)

Year	Area	Sampling period	Habitat and plant species					
			Crop		Non crop			
			То	Eg	То	Sn	Ng	Total
2008	NE	22/7-18/10	22			1		23
	CE	15/6–7/9	32					32
2009	NE	21/4-25/9	38	4		2		44
	CE	12/6-7/11	59					59
	SE	27/10-1/12				6		6
2010	NE	2/7-5/10	9	2		2		13
	SE	10/2-23/12			7	13	4	24
2011	NE	23/6-24/10	25			1		26
	SE	14/1–6/7			2	19		21

Plant species were tomato (To), eggplant (Eg), Solanum nigrum (Sn) and Nicotiana glauca (Ng)

IPM or organically managed and included mainly tomato (*Lycopersicon esculentum* Mill.), but also some eggplant (*Solanum melongena* L.), grown in open field or in non insect-screened greenhouses. In the CE, all naturally infested samples were from tomato crops (either open field or greenhouses) mostly managed with insecticides. In the SE, the samples were from non-crop plants (*S. nigrum*, *Nicotiana glauca* Graham and tomato plants found in natural habitats). In this area, crops were not sampled since most of them were under insectscreened greenhouses and insecticide managed.

In commercial tomato crops, at least 50 tomato leaflets infested by *T. absoluta* were randomly collected per sample. Eggplant crops and non-crop plants (*S. nigrum*, *N. glauca* and non-cultivated tomato) were surveyed during 15 min and leaves with *T. absoluta* galleries were collected. Each sample was placed in a plastic bag and refrigerated at 5–10 °C for less than one week until processed in the laboratory. Insects other than *T. absoluta* were carefully removed and pieces of leaves with *T. absoluta* galleries were cut and enclosed in aerated plastic cages at room temperature (20–25 °C and 50–80 % RH) for four weeks. Cages were reviewed twice a week and parasitoid adults were collected and kept in 70 % alcohol for identification. Parasitoid species were morphologically identified by A. Ribes and M.J. Verdú.

Sentinel plants

To increase the recruitment of parasitoid species that are able to parasitize T. absoluta, sentinel plants were used in the three tomato production areas mentioned above. Samplings in the CE area were all conducted in the locality of Moncada (Valencia) (39.6°N 0.4°W, 33 m altitude) in four different scenarios: (i) the vicinity of a tomato crop, (ii) a Mediterranean pine forest, (iii) a citrus orchard with a multi-species spontaneous ground cover and (iv) a citrus orchard with Festuca arundinacea Schreber (Poaceae) ground cover. In the NE area, samplings were all conducted in the locality of Cabrils (41.5°N 0.6E, 81 m altitude), in an area close to horticultural crops and to a flowering margin planted to enhance the presence of natural enemies. In the SE, samplings with sentinel plants were all performed in the locality of La Mojonera (36.47°N 2.42°W, 142 m altitude) in a very intensive greenhouse vegetable production area. Table 2 summarizes the number of samples and the survey periods. Each sample consisted of three or four tomato plants (var. Bond F1, De Ruiter

Table 2 Number of samples of sentinel tomato plants in three tomato production areas located in the north (NE), central (CE) and south (SE) part of the Mediterranean coast of Spain (see coordinates in the text)

Year	Area	Sampling period	Eggs	Larvae
2009	NE	July-December	20	-
	CE	August-December	68 ^a	_
2010	NE	July-November	21	20
	CE	July-September	44 ^a	_
	SE	February-December	11	_
	SE	April–December	_	9
2011	NE	July-November	_	21

Each sample comprised 3-4 plants infested either with *T. absoluta* eggs or larvae and exposed for 72 h

^a Four samples in four different habitat scenarios per week. See text for details

Seeds) *circa* 20–30 cm high and previously infested with either eggs (40–200 eggs per plant) or second–third instars of *T. absoluta* larvae (100–200 larvae per plant). Sentinel plants were exposed weekly for 72 h. Plants were placed on the top of a brick inside a plastic tray full of water to avoid access by non-flying arthropod predators. After 72 h, plants were returned to the laboratory and eggs and larvae were confined in aerated cages at room temperature. Cages were prepared and check and parasitoids were collected and identified as described in the previous section.

Statistical analysis

The proportion of naturally infested samples yielding at least one parasitoid and the number of parasitoid species found repeatedly across the years were analyzed using GLM with binomial errors and logit link function. The model included the term year as the independent variable and its significance was tested by comparing it with the null model. Proportions of samples with parasitism from crops in the NE area and from non-crop habitats in the SE area were analyzed separately. The statistical analyses were performed using R (R Development Core Team, 2008).

Results

Parasitoids emerged from 58 of these, which represented 23 % of the total number of samples (Table 3). The proportion of crop samples from the NE area yielding at least one parasitoid significantly increased across years ($\chi^2 = 9.087$; df = 3; P = 0.028) (Fig. 1a). Conversely, proportion of non-crop samples from the SE area yielding parasitism was not different among years ($\chi^2 = 0.136$; df = 2; P = 0.934) (Fig. 1b). Adult parasitoids were obtained from tomato, eggplant and *S. nigrum* but not from the samples of *N. glauca*. A high frequency of parasitism occurred on eggplant, non-cultivated tomato and *S. nigrum*.

Table 3 summarizes *T. absoluta* parasitoid species found in the surveyed areas from 2008 to 2011. In total, 20 different species were reared from larva/ pupae belonging to five different families, but five of these have only been identified to genus level. Larval parasitoids include nine Eulophidae, seven Braconidae, two Ichneumonidae, one Pteromalidae and one Chalcididae species. Egg parasitoids of *T. absoluta* were recruited from sentinel tomato plants in the three surveyed areas and all of them belong to *Trichogramma* genus.

Seven out of the 20 species of larval/pupal parasitoids were found more than one year. Among these, five belong to Eulophidae and two to Braconidae families: *Necremnus* sp. nr. *artynes* (Walker), *Stenomesius* cf. *japonicus* (Ashmead), *Necremnus* sp., *N. formosa* and *D. isaea* (Hymenoptera: Eulophidae) and *Choeras semele* (Nixon) and *Apanteles* sp. (Hymenoptera: Braconidae). Number of species repeatedly reared from *T. absoluta* increased with years ($\chi^2 = 8.101$; df = 3; *P* = 0.044) and were two in 2008, three in 2009, and six in 2010 and 2011. The remaining thirteen species were only found one year and in one area.

Necremnus sp. nr. artynes was the only species that was found in all four years and in all the tomato production areas included in the survey. It was found in both cultivated and non-cultivated plants and in the three different host plant species: tomato, eggplant and *S. nigrum*. In addition, another species of Necremnus yet to be identified was found in tomato in the NE and SE areas. Stenomesius cf. japonicus was found in three out of the four years of sampling, only in the NE area and in all plant species: tomato, eggplant and *S. nigrum*. Neochrysocharis formosa was found in the two last years of the survey, in two areas and on crop and non-crop plants of tomato and on *S. nigrum*. In the SE area, *D. isaea* was detected parasitizing *T. absoluta*

 Table 3 Parasitoid species of *Tuta absoluta* found in the surveyed areas located in the north (NE), central (CE) and south (SE) parts of the Spanish Mediterranean coast from 2008 to 2011

	Year and area				Habitat and plant species	
Species	2008	2009	2010	2011	Crop	Non crop
LARVAL/PUPAL PARASITOIDS						
Eulophidae						
*Necremnus sp. nr. artynes (Walker, 1839)	NE^1 , $CE^{2,3}$	NE, CE	NE, SE ^{4,11}	NE, SE	$\bigcirc \blacklozenge$	Ο×
*Stenomesius cf. japonicus (Ashmead, 1904)	NE ⁵	NE	NE		$\bigcirc \blacklozenge$	⊙×
*Necremnus sp.		NE ⁶		SE ⁷	0	•
*Neochrysocharis formosa (Westwood, 1833)			NE ⁸ , SE ⁴	NE, SE	0	• ×
*Diglyphus isaea (Walker, 1838)			SE 4,9	SE		Ο×
Diglyphus crassinervis Erdös, 1958				NE 10	0	
Elasmus phthorimaeae Ferriere, 1947			SE ^{4,7}			⊙×
Pnigalio cristatus (Ratzeburg, 1848)			NE ⁸			•
Pnigalio soemius (Walker, 1839)		NE ⁶			0	
Braconidae						
*Apanteles sp.			SE ^{4,11}	SE		×
*Choeras semele (Nixon 1965)			SE ¹¹	SE		×
Cotesia sp.	NE ¹²				0	
Dolichogenidea litae (Nixon, 1972)	NE ^{5,13}				0	×
Bracon (Habrobracon) sp. nr. nigricans (Szépligeti, 1901)			NE ⁸			•
Chelonus sp.		SE ^{11,14}				×
Diolcogaster sp.				SE15		×
Ichneumonidae						
Temelucha anatolica (Sedivy, 1959)	NE ¹⁶				0	
Zoophthorus macrops Bordera & Horstmann, 1995			NE ⁶		0	
Chalcididae						
Hockeria unicolor Walker, 1834			NE ⁸			•
Pteromalidae						
Pteromalus semotus (Walker, 1834)				NE ⁸		•
Egg parasitoids						
Trichogramatidae						
Trichograma sp.		NE ⁸ , CE ¹⁷	NE, SE ⁹			•

Superscripts in the area acronyms indicate the locality in which this association was found for the first time in Spain: 1, Santa Susanna; 2, Alcalà de Xivert; 3, Meliana; 4, La Cañada; 5, Amposta; 6, Blanes; 7, Nijar; 8, Cabrils; 9, La Mojonera; 10, Cabrera de Mar; 11, Dalías; 12, Castellvell; 13, Argentona; 14, Cabo de Gata; 15, El Ejido; 16, Vinyols; 17, Moncada. Symbols indicate plants where species were found: \bigcirc tomato crop, \bullet tomato sentinel plant, \odot non-cultivated tomato, \bullet eggplant and \times *S. nigrum*. Asterisk indicates the parasitoid species that were found in more than one year

larvae on tomato and *S. nigrum* in two consecutive years. *Choeras semele* and *Apanteles* sp., were found in the two last years of sampling, but only on *S. nigrum*. The other 13 species were only found in a single year and in one of the sampled areas.

Regarding the habitat, ten different species emerged from tomato and eggplant crop samples (n = 192) and 15 from non-crop samples (n = 107), including *S. nigrum* and non-cultivated tomato (spontaneous seedlings in wild habitats and sentinel plants). Only four species were common to both habitats: *N.* sp. nr. *artynes*, *S.* cf. *japonicus*, *Necremnus* sp. and *N. formosa*. Overall, 16 species were reared from tomato (both cultivated and non-cultivated), ten species from

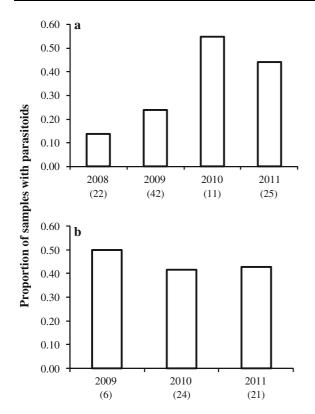


Fig. 1 Proportion of *T. absoluta* samples collected in crop **a** or in non-crop **b** habitats yielding at least one parasitoid. Numbers in brackets indicate the number of samples per year

S. nigrum and two from eggplant. Four parasitoid species were only recruited on *S. nigrum: C. semele*, *Apanteles* sp., *Chelonus* sp. and *Diolcogaster* sp. (Hymenoptera: Braconidae).

Egg parasitoids of *T. absoluta* were recruited from sentinel tomato plants in the three surveyed areas (Table 3). In the NE area, egg parasitoids emerged from eight samples out of 20 in 2009 and from three out of 20 in 2010. In the CE area, one single egg parasitoid was obtained from a sentinel plant placed in the pine forest in 2009, and none during 2010. In the SE, three individuals emerged from one sentinel plant in 2010. All individuals that emerged from the eggs were identified to belong to the *Trichogramma* genus.

Discussion

one year indicating that are repeatedly using this pest as host. Out of these, two were already present in the first sampling year and six the last two, which indicates that the species richness of T. absoluta larval/pupal parasitoids in the tomato production coastal areas of Spain increased with time. For C. semele this paper is the first report of a host-parasitoid association with T. absoluta. We have also found other 13 larval/pupal parasitoid species occasionally parasitizing T. absoluta. Six of those were first reported to be able to parasitize this lepidopteran pest in this paper. These species are. Elasmus phthorimaeae Ferriere and Diglyphus crassinervis Erdös (Hymenoptera: Eulophidae), Dolichogenidea litae (Nixon) (Hymenoptera: Braconidae), Temelucha anatolica (Sedivy) and Zoophthorus macrops (Bordera & Horstmann) (Hymenoptera: Ichneumonidae) and Pteromalus semotus (Walker) (Hymenoptera: Pteromalidae).

Other surveys have been conducted in the Mediterranean looking for associations between larval/ pupal parasitoids and T. absoluta. In Italy, where T. absoluta was first detected in 2008, a similar number of species has been found (Ferracini et al. 2012; Loni et al. 2011; Zappalà et al. 2012). Lower parasitoid richness has been detected in other areas: nine species in Turkey (Doğanlar and Yiğit 2011), six species in Algeria (Boualem et al. 2012) and one species in Jordan (Al-Jboory et al. 2012). Overall, from 2008 to the present date, 41 different taxa of larval/pupal parasitoids have been detected exploiting T. absoluta as a host in the Mediterranean. These parasitoids comprise 19 Eulophidae, 12 Braconidae, six Ichneumonidae, two Pteromalidae and two Chalcididae. Cornell and Hawkins (1993) observed that the parasitoid richness of many herbivores in an invaded area tends to increase with herbivore residence time. They suggest that it occurs for at least two reasons. First, the period of adjustment of the parasitoid behavior or phenology to the new host may be different for different parasitoid species. Second, the geographical spread of the host may favor encounters with new parasitoids. Unexpectedly, the guild of larval/pupal parasitoids of T. absoluta reported in the Mediterranean basin is larger than that found in its area of origin, which includes 29 taxa (Desneux et al. 2010; Luna et al. 2012). This may be related to an actual higher species richness attacking this niche in the Mediterranean than in South America. It may also be a consequence of insecticide use being the main tactic employed against the pest in the area of origin (Guedes and Picanço 2012) and an increasing use of biological control in the Mediterranean (Arnó and Gabarra 2011; González-Cabrera et al. 2011; Mollá et al. 2011) that has probably contributed to providing the parasitoids with suitable habitats where to reproduce. The same argument on the influence of crop management practices on the parasitoid richness may explain results obtained in our survey. Fifteen different species of T. absoluta parasitoids were reared from 107 samples from non-crop habitats whereas only ten were recovered from the 191 commercial crops samples. Besides, the proportion of samples yielding at least one parasitoid in the non-crop habitat was close to 50 % during the three years of the survey but increased from 14 % to 44 % in the crop samples from 2008 to 2011. Although, in the NE, the fields were managed according to IPM programs based on biological control, higher knowledge of the effectiveness of insecticides made possible a better conservation of naturally existing parasitoids (Arnó and Gabarra 2011; González-Cabrera et al. 2011; Mollá et al. 2011).

Among the larval/pupal parasitoids that we have found repeatedly on T. absoluta, N. sp. nr. artynes was found all the years and with a wider distribution. This species is currently under taxonomic revision and may actually consist of a species complex (Ferracini et al. 2012; Zappalà et al. 2012). Necremnus sp. nr. artynes or *N. artynes* was one of the earliest parasitoids to be reported on T. absoluta in Europe, and has been recorded in Italy, Spain and Algeria (Boualem et al. 2012; Ferracini et al. 2012; Urbaneja et al. 2012; Zappalà et al. 2012). Zappalà et al. (2012) found N. nr. artynes spontaneously parasitizing T. absoluta from November to May, which indicates that it may overwinter in warm climates. The estimated intrinsic rate of increase of N. artynes is higher than that of T. absoluta on tomato, indicating its potential to control this pest (Calvo et al. 2013). Stenomesius cf. japonicus has been previously reported as a T. absoluta parasitoid in Spain (Urbaneja et al. 2012). The form that has been found parasitizing T. absoluta shows differences in morphology from the type description. Hence a further revision is needed for this species. Neochrysocharis formosa was found parasitizing T. absoluta in Spain and Italy (Ferracini et al. 2012; Lara et al. 2010; Zappalà et al. 2012). Recently, it has been recorded in Argentina and is the only species recruited in the Mediterranean that has also been reported in the T. absoluta origin area (Luna et al. 2011). It is considered to be one of the parasitoids that controlled the invasion of Liriomyza spp. in Europe and in the Mediterranean (Gabarra and Besri 1999; Nicoli 1997) and it has been also reared from Phyllocnistis citrella Stainton (Lepidoptera: Gracillariidae), an invasive pest of citrus crops in the Mediterranean Basin (Massa et al. 2001; Urbaneja et al. 2000). Finally, D. isaea has also been recorded on T. absoluta in Algeria (Boualem et al. 2012). Despite the fact that the parasitism levels of D. isaea on Liriomyza spp. are very high in the coastal areas of Spain (Gabarra and Besri 1999), it was only found on T. absoluta during the last two years of the survey and only in one area, suggesting that T. absoluta is a non-preferred host.

In our survey we found other four Palearctic species that have also been reported in other Mediterranean areas on *T. absoluta* and therefore thay may be considered part of its guild of parasitoids in the Mediterranean area: B. sp. nr. nigricans in Italy and Jordan, P. cristatus and Pnigalio soemius (Walker) (Hymenoptera: Eulophidae) complex found in Italy and Turkey, and Hockeria unicolor Walker (Hymenopera: Chalcididae) found in Turkey (Al-Jboory et al. 2012; Doğanlar and Yiğit 2011; Ferracini et al. 2012; Urbaneja et al. 2012; Zappalà et al. 2012). The form of B. nigricans found in Spain is currently under revision (K. Achterberg, personal communication) and the broad concept of P. soemius may include cryptic species that are also currently under revision (Bernardo et al. 2008; Gebiola et al. 2010).

All egg parasitoids that we found during our survey belonged to the Trichogramma genus (Table 3), but rates of parasitism were very low (only 8 % of samples). In the same areas egg parasitism rates of Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae) and C. chalcites in tomato crops by Trichogramma spp. were up to 80 % (Gabarra, Arnó & Riudavets, unpublished). This large difference in parasitism rates suggests that the Trichogramma species present in the area have probably little affinity for T. absoluta or for the association of T.absoluta/ tomato as host/host-plant complex (Chailleux et al. 2012). Zappalà et al. (2012) also found several Trichogramma spp. parasitizing T. absoluta in South Italy, and consider there may be up to five species associated with this pest. In South America, T. absoluta has a wide guild of egg parasitoids with more than 12 Trichogrammatidae, four Encyrtidae and one Eupelmidae species/genera (Desneux et al. 2010). This higher egg parasitoid richness found in South America may be due to at least two reasons. The first is a more intensive monitoring of *T. absoluta* egg parasitism since, in this region, biological control of the pest has mainly been attempted using egg parasitoids (Guedes and Picanço 2012). The second may be that *Trichogramma* species are generally scarcer in the Mediterranean than in South America (Sumer et al. 2009; Zucchi et al. 2010).

Currently, integrated pest management programs for tomato crops in the Mediterranean are based on the use of the predators *N. tenuis* and *M. pygmaeus*, which effectively control *T. absoluta* (Urbaneja et al. 2012). Our survey indicated that *N.* sp. nr. *artynes*, *N. formosa* and *S.* cf. *japonicus* are potentially interesting candidates as biocontrol agents. Since these parasitoids target larval/pupal stages and the predators are basically *T. absoluta* egg consumers, their activity may be complementary and improve pest control.

Since species richness and proportion of samples with parasitoids were high in non-cultivated plants, non-crop habitats may be an important source of natural enemies that enhance conservation biological control (Schmidt et al. 2004). One of the measures that may be implemented is the use of insectary plants to enhance the abundance of predators and parasitoids in vegetable crops (Alomar et al. 2006; Arnó et al. 2012; Burgio et al. 2007).

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