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Morphometric and molecular analysis of the *Encarsia inaron* species-group (Hymenoptera: Aphelinidae), parasitoids of whiteflies (Hemiptera: Aleyrodidae)

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Abstract

Several series of host-reared specimens of an *Encarsia* species, initially thought to be the cosmopolitan *Encarsia inaron* (Walker), were collected in the Azores Islands (Portugal). Subsequent morphometric analysis supported the presence of two species: *E. inaron* and a new species, described herein as *Encarsia estrellae* Manzari & Polaszek **sp. n.** *Encarsia estrellae* was reared from *Aleyrodes singularis* Danzig, *A.* ?*singularis*, and *Bemisia* sp. *afer*-group on several host plants. In addition, the D2 region of the 28S rDNA gene was sequenced in eight individuals belonging to these species, as well as single representatives of two closely related and one distantly related species. Phylogenetic analysis of these DNA sequences, together with 23 additional *Encarsia* sequences retrieved from the European Molecular Biology Laboratory (EMBL) and GenBank databases, further supported the specific status of *E. estrellae*, and the placement of *E. dichroa* (Mercet) in the *E. inaron* species-group. Additionally, *E. inaron* is redescribed and some taxonomic problems in the *E. inaron* species-group are discussed.

Introduction

Encarsia Förster (Hymenoptera: Aphelinidae) is a taxonomically difficult genus, which currently contains about 280 described species (Polaszek *et al*., 1999). *Encarsia* species are mostly primary parasitoids of whiteflies (Hemiptera: Aleyrodidae) and armoured scale insects (Hemiptera: Diaspididae) – on which they are important biological control agents – (Huang & Polaszek, 1998), but four species are parasitoids of aphids (Hemiptera: Aphididae: Hormaphidinae) (Evans *et al*., 1995) and a few species parasitize the eggs of Lepidoptera (Polaszek, 1991).

Although recent papers (Viggiani, 1985, 1987; Hayat, 1989, 1998; Polaszek *et al*., 1992, 1999; Evans & Polaszek, 1998; Huang & Polaszek, 1998) have contributed to the identification of these parasitoids, reliable identification is still difficult for many species. This is for several reasons, e.g. their small size, diversity, and the existence of complexes of morphologically indistinguishable or hard to distinguish species (Polaszek *et al*., 1999). The assignment of species-groups to *Encarsia* and the placement of the species within species-groups is also problematic, and a revision of the genus on a world basis is needed (Hayat, 1998). Approximately 29 species-groups are currently recognized

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within *Encarsia* but few of these groups can be recognized by discrete morphological characters. Indeed, for this reason, some species have been included in different groups by different authors (Viggiani & Mazzone, 1979; Heraty & Polaszek, 2000).

In September 1998 and June 2000 the second author and co-workers reared several series of an *Encarsia* species from several localities, and from different hosts and host plants, in the Azores Islands. After slide-mounting and examination with compound microscopy these were found to be morphologically extremely close to, if not the same as, the cosmopolitan *E. inaron* (Walker). Specimens were reared from *Aleyrodes singularis* Danzig, *A.* ?*singularis* (see note below under 'hosts'), and *Bemisia* sp. *afer*-group whiteflies (Hemiptera: Aleyrodidae). In this study we attempt to obtain objective views of species boundaries using principal component analysis (PCA) and canonical discriminant analysis (CDA) (Blackman & Paterson, 1986; Narusis, 1993; Chishti & Quicke, 1996; Añez *et al.*, 1997; Azidah *et al.*, 2000; Heraty & Polaszek, 2000). DNA sequence data were also used to test our findings and explore the support for existing species-groups. The D2 region of the rDNA gene was chosen for compatibility with the earlier study of Babcock *et al.* (2001). The *E. inaron* species-group currently contains 16 described species (http://cache.ucr.edu/~heraty/Encarsia.cat.pdf). In this catalogue Heraty & Woolley do not include *E. dichroa* (Mercet) in the *E. inaron* species-group, although Hayat (1998) does so. Our analysis of the *E. inaron* species-group also includes the first sequences for *E. dichroa* and *E.* sp. near *azimi* Hayat, from Cameroon. A morphological diagnosis of the group is provided by Hayat (1998), who, for Indian species, gives the following combination of characters: ovipositor shorter than mid tibia and basitarsus combined, mid tibial spur less than half-length of basitarsus, and clava 2-segmented. The applicability of this diagnosis was therefore tested. Finally, the new material is described as *E. estrellae* Manzari & Polaszek **sp. n.** and *E. inaron* is redescribed.

Materials and methods

Morphometric analysis

One hundred specimens of *E. inaron*, 59 females and 41 males, including the holotype and paratypes of *E. borealis* Huldeń (synonymized by Huang & Polaszek, 1998), and 11 specimens of *E. estrellae* (5 females and 6 males) were each measured using slide-mounted preparations and an ocular graticule. Forty-six variables (25 continuous and 21 discrete) were recorded for females, and 37 variables (22 continuous and 15 discrete) were recorded for males. Morphological terminology follows that of Hayat (1989) except the use of 'mesosoma' instead of 'thorax'. Material examined (table 1) belongs to the following collections: The Natural History Museum, London, UK (BMNH); Museo Nacional de Ciencas Naturales, Madrid, Spain (MNCN); Zoological Museum of the University of Helsinki, Finland (MZH). Continuous and discrete variables were analysed both separately and in combination. Measurements (continuous variables) used for PCA and CDA are illustrated in fig. 1 and listed in the legend.

Discrete variables were as follows: numbers of longitudinal sensilla (rhinaria) of the antennal flagellomeres (6 variables) (females only), numbers of setae on both left and right sides of mid lobe (2 variables) and side lobes (2 variables) of the mesoscutum, numbers of setae on both left and right sides of tergites 1–4 (8 variables) and all setae on tergites 5–7 (3 variables). Males and females were treated separately and the analyses were all carried out on untransformed data (Quartau, 1982; Blackman & Spence, 1994) based on correlation matrices using the statistical package SPSS (Narusis, 1993). Analyses of log-transformed data were also carried out and gave virtually the same results (not presented). Cases with missing values were treated in two different ways: replaced with the mean or excluded from the analysis. Since results of treating missing data in the two ways were almost the same, only the latter is presented.

DNA extraction, amplification and sequencing

Adults were killed directly in 70–95% ethanol. DNA was extracted from single specimens using the DNeasy Tissue Kit (Qiagen, Crawley, UK) with elution into 30 μl distilled water. Standard 50 μl polymerase chain reaction (PCR) was then carried out in a GeneAmp 9600 thermal cycler containing 1.0 μl DNA extract, 5 μl *Taq* buffer (1.5 mM $MgCl_2$), 1.5 U *Taq* polymerase (Roche), 10 nmol dNTPs (Amersham Pharmacia Biotech; APB, Amersham, UK) and 20 pmol of each primer. The D2 region of 28S rDNA was amplified using the following primers: forward 5′-GCG AAC AAG TAC CGT GAG GG-3′; reverse 5′-TAG TTC ACC ATC TTT CGG GTC-3′ (note, this product includes the smaller D3 region, which was not used in our analyses). GFX gel band purification (APB) was used to clean all PCR products, which were then sequenced in both directions with the same primers using *Big Dye* terminators at half recommended volumes on an ABI Prism 3700 automated sequencer. PCR conditions were 35 cycles of 95°C denaturation (30 s), 45°C annealing (30 s) and 72°C extension (1 min) with an initial denaturation for 2 min and a final extension for 4 min. The 28S-D2 locus was sequenced for four individuals of *E. estrellae* from a total of three localities in the Azores Islands, and four individuals of *E. inaron* from UK, Japan (two individuals) and India. Single individuals of two other probable members of the *E. inaron* species-group, *E. dichroa* and *E.* sp. near *azimi* (Cameroon) were also sequenced, plus a third unrelated species (*E. tricolor* Förster). Sequences have been deposited in the European Molecular Biology Laboratory (EMBL) and GenBank databases under accession numbers AJ305294–AJ305302 (table 2).

Phylogenetic analysis

Amplified D2 regions were between 454 and 457 bp in length. The sequences obtained, together with 26 additional ones retrieved from EMBL/GenBank (from Babcock *et al.*, 2001) including representatives of three related genera, were aligned by eye and also using the Clustal X programme with four sets of gap opening and gap extension penalties: 20:10, 15:6.66 (default), 10:5 and 5:1 (the default downweighting of transitions by 0.5 was used in all). Maximum parsimony analysis (MP) was then performed on the five multiple alignments using PAUP* (Swofford, 1998). Heuristic searches were carried out treating gaps as both missing data and informative with 1000 random additions followed by branch swapping using tree-bisection-reconnection (TBR) and unlimited maxtrees. In each alignment the bootstrap

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Table 1. Geographical and biological information of *Encarsia* specimens used in the morphometric analysis.

Species	Locality	n	Host species	Host plant
E. inaron	Afghanistan	2	Unknown	*Rosa* sp.
	Austria	1	Unknown	Unknown
	Bulgaria	1	*Siphoninus phillyreae* (Haliday)	*Crataegus* sp.
	Egypt	4	*Trialeurodes vaporariorum* (Westwood)	*Ricinus communis*
	Egypt	3	*Siphoninus phillyreae* (Haliday)	*Punica granatum*
	Egypt	2	Unknown	Unknown
	England	6	*Aleyrodes proletella* L.	*Brassica oleracea*
	England	3	*Aleyrodes proletella* L.	*Urtica dioica*
	England	2	*Aleyrodes proletella* L.	Unknown
	England	1	*Aleyrodes proletella* L.	Crucifer
	England	3	*Tetraleurodes* ?*hederae* Goux	*Hedera helix*
	England	1	*Pulvinaria vitis* (?misidentification)	*Vitis* sp.
	England	1	*Trialeurodes vaporariorum* (Westwood)	*Solanum* sp.
	England	1	Unknown	Unknown
	Finland	2	*Pealius quercus* (Signoret)	Unknown
	France	1	*Aleyrodes lonicerae* Walker	*Thalictrum minus*
	France	1	*Aleyrodes proletella* L.	*Chelidonium majus*
	India	1	Unknown	Unknown
	India	1	*Siphoninus phillyreae* (Haliday)	Unknown
	Iran	6	Unknown	Unknown
	Iran	3	*Siphoninus phillyreae* (Haliday)	Unknown
	Israel	2	Unknown	Unknown
	Italy	7	*Siphoninus phillyreae* (Haliday)	*Fraxinus* sp.
	Italy	3	*Pealius azaleae* (Baker & Moles)	Unknown
	Italy	1	Unknown	*Robinia* sp.
	Jordan	5	*Aleyrodes singularis* Danzig	Unknown
	Jordan	1	Unknown	*Citrus* sp.
	New Zealand	2	Unknown	Unknown
	New Zealand	2	*Siphoninus phillyreae* (Haliday)	Unknown
	Pakistan	7	*Bemisia tabaci* (Gennadius)	*Gossypium hirsutum*
	Pakistan	1	Unknown	Unknown
	South Africa	2	Unknown	*Lycopersicon esculentum*
	South Africa	1	Unknown	Unknown
	Syria	1	Unknown	*P. granatum*
	Syria	4	*Aleyrodes singularis* Danzig	*Lactuca* sp.
	Thailand	1	Unknown	Unknown
	Turkey	4	*Siphoninus phillyreae* (Haliday)	*Pyrus communis*
	Turkey	4	*Aleyrodes proletella* L.	*B. oleracea*
	Turkey	3	*Aleyrodes* sp.	*Sonchus* sp.
	Turkey	1	*Aleurothrixus floccosus* (Maskell)	Unknown
	Venezuela	2	*Siphoninus phillyreae* (Haliday)	*P. granatum*
E. estrellae	Azores (Portugal)	1	*Bemisia* sp.	*Viburnum tinus subcordatum*
	Azores (Portugal)	2	*Bemisia* sp.	*H. helix canariensis*
	Azores (Portugal)	3	*Bemisia afer*-group	*Ilex perado azorica*
	Azores (Portugal)	2	*Aleyrodes* ?*singularis* Danzig	*Lysimachia nemorum*
	Azores (Portugal)	3	*Aleyrodes singularis* Danzig	*L. nemorum*

support for individual branches was found using 100 pseudoreplicates each of 100 random additions (Felsenstein, 1985).

Results

Principal component analysis

Five components were extracted based on continuous variables for each analysis and biplots of all pair-wise combinations of these examined. Eigenvalues and weights for first two principal components are presented in table 3. Figure 2a,b shows scatter diagrams of female and male *Encarsia* with respect to first and second principal components obtained using continuous variables. In both sexes, the specimens can be seen to fall into two groups, with no overlap. Females and males of the two *Encarsia* species were projected on the first two principal components (PC-I and PC-II), which together accounted for 74.5% and 77.8% of the overall variance respectively. Length of marginal fringe and distance between placoid sensilla had the lowest contributions, and length of meta tibia, length of scutellum and length of fore wing had the highest influences on PC-I for male *Encarsia* (see table 3). Other variables had approximately equal contributions. For female *Encarsia*, length of third valvula, distance between placoid sensilla and length of marginal fringe had the lowest contributions, and length of funicle and length of flagellomeres 3 and 4 had the highest influences on PC-I (see table 3). As with males, other variables had approximately equal contributions.

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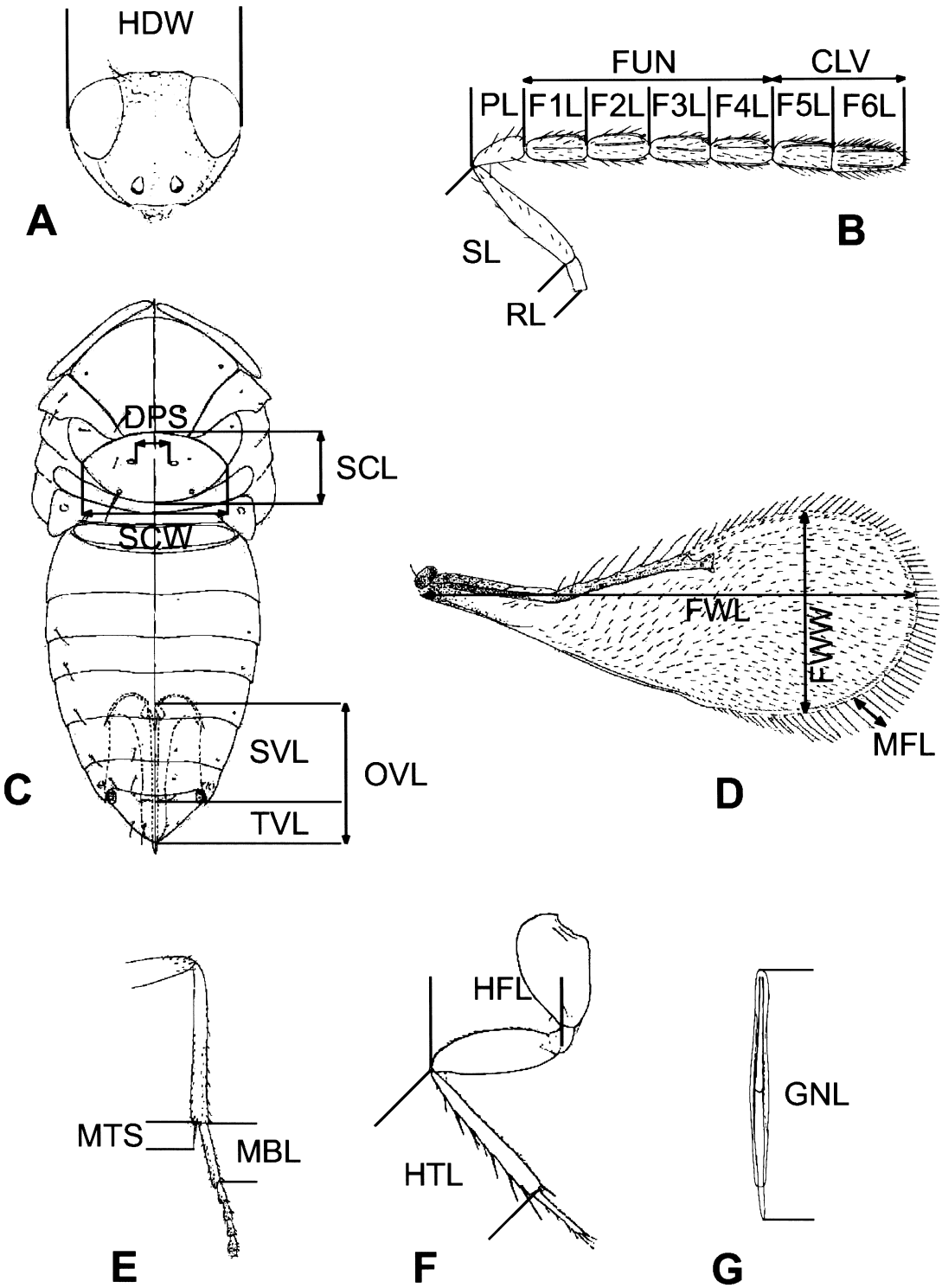

Fig. 1. Measurements (continuous variables) used for principal component analysis and canonical discriminant analysis. A, head; B, antenna; C, mesosoma and gaster; D, fore wing; E, mid leg; F, hind leg; G, male genitalia. CLV, clava length; DPS, distance between placoid sensilla; F1L, flagellomere 1 length; F2L, flagellomere 2 length; F3L, flagellomere 3 length; F4L, flagellomere 4 length; F5L, flagellomere 5 length; F6L, flagellomere 6 length; FUN, funicle length; FWL, fore wing length; FWW, fore wing width; GNL, male genitalia length; HDW, head width; HFL, hind femur length; HTL, hind tibia length; MBL, meso-basitarsus length; MFL, marginal fringe length; MTS, mesotibial spur length; OVL, ovipositor length; PL, pedicel length; RL, radicle length; SCL, scutellum length; SCW, scutellum width; SL, scape length; SVL, second valvifer length; TVL, third valvula length. A, F and G redrawn from Hayat (1998); B–E redrawn from Polaszek *et al*. (1999).

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Table 2. Specimens of *Encarsia* used in the molecular phylogenetic analysis and their geographical and biological information.

Taxa	Locality	Host	EMBL/GenBank accession number
E. sp. near *azimi* Hayat	Cameroon, Mbalmayo	Unknown	AJ305294
E. estrellae sp.n. (az2)	Azores Islands, São Miguel	*Bemisia* sp. on *Viburnum* sp.	AJ305297
E. estrellae sp.n. (az3a)	Azores Islands, Pico, Chão Vert	*Bemisia* sp. on *Rubia peregrina*	AJ305298
E. estrellae sp.n. (az3b)	Azores Islands, Pico, Lagoa do Caiado	*Bemisia* sp. on *R. peregrina*	AJ305299
E. estrellae sp.n. (az4)	Azores Islands, Pico, Lagoa do Caiado	*Bemisia* sp. on *Euphorbia stygiana*	AJ305300
E. dichroa (Mercet)	Spain, Granada	*Siphoninus* sp.	AJ305301
E. inaron (Walker)	England, London	*S. phillyreae* (Haliday)	AJ305295
**E. inaron* (Walker)	Japan, Sanyocho	*S. phillyreae* (Haliday) on *Punica granatum*	–
**E. inaron* (Walker)	Japan, Yamaguchi	*S. phillyreae* (Haliday) on *P. granatum*	–
E. inaron (Walker)	India, Tamil Nadu, Arrupukottai	Unknown	AJ305296
E. tricolor Förster	Azores Islands, São Miguel	Unknown	AJ305302

* These two sequences and the sequence of *E. inaron* from England are identical, so only the latter sequence was deposited in EMBL/GenBank databases.

Table 3. Eigenvalues and weights for first two principal components extracted based on continuous variables for females and males.

Variable	Principal components			
	I	II	I	II
	Females		Males	
Eigenvalues	16.76	1.88	15.84	1.28
HDW	0.0453	0.1961	0.0477	−0.2162
RL	0.0369	0.2701	0.0411	−0.0933
SL	0.0533	0.0971	0.0578	−0.0669
PL	0.0492	0.1338	0.0484	0.0922
F1L	0.0544	0.0155	0.0585	0.1067
F2L	0.0544	−0.0350	0.0581	0.1343
F3L	0.0569	−0.0211	0.0592	0.0681
F4L	0.0569	−0.0694	0.0590	−0.0002
FUN (females only)	0.0580	−0.0265	–	–
F5L	0.0536	−0.1420	0.0583	−0.1059
F6L	0.0509	−0.1781	0.0542	−0.2255
CLV	0.0541	−0.1656	0.0581	−0.1681
FWL	0.0551	−0.0011	0.0596	0.1209
FWW	0.0492	0.0713	0.0574	−0.0081
MFL	0.0346	−0.3034	0.0292	0.6468
SCL	0.0530	0.0871	0.0597	−0.1009
SCW	0.0483	0.1128	0.0545	−0.0811
DPS	0.0313	−0.0828	0.0305	−0.2064
HFL	0.0480	0.0648	0.0544	−0.0149
HTL	0.0539	0.1145	0.0617	−0.0247
MBL	0.0501	−0.0401	0.0543	−0.0546
MTS	0.0415	−0.2065	0.0510	0.1162
SVL (females only)	0.0418	−0.1375	–	–
TVL (females only)	0.0295	0.3189	–	–
OVL (females only)	0.0451	−0.0005	–	–
GNL (males only)	–	–	0.0504	0.2805

See fig. 1 for abbreviations.

Canonical discriminant analysis

Separation between the putative species was more marked using canonical discriminant analysis of continuous variables, and combined continuous and discrete variables (Chishti & Quicke, 1996). Based on continuous variables, females and males were projected on the first and second canonical discriminant function (CD-I and CD-II), which together accounted for 73.4% and 72.6% of the original variance respectively (fig. 2c,d). Analysis based on combined continuous and discrete variables for both sexes gave no more separation than analysis based on continuous variables only (fig. 2e,f).

Phylogenetic relationships

Length of multiple alignments was 493–495, with 179–188 parsimony informative positions (gaps treated as informative). Maximum parsimony (MP) analyses of the 37 DNA sequences gave a total of 221 most-parsimonious trees (MPTs) from the five alignments and one of them is shown

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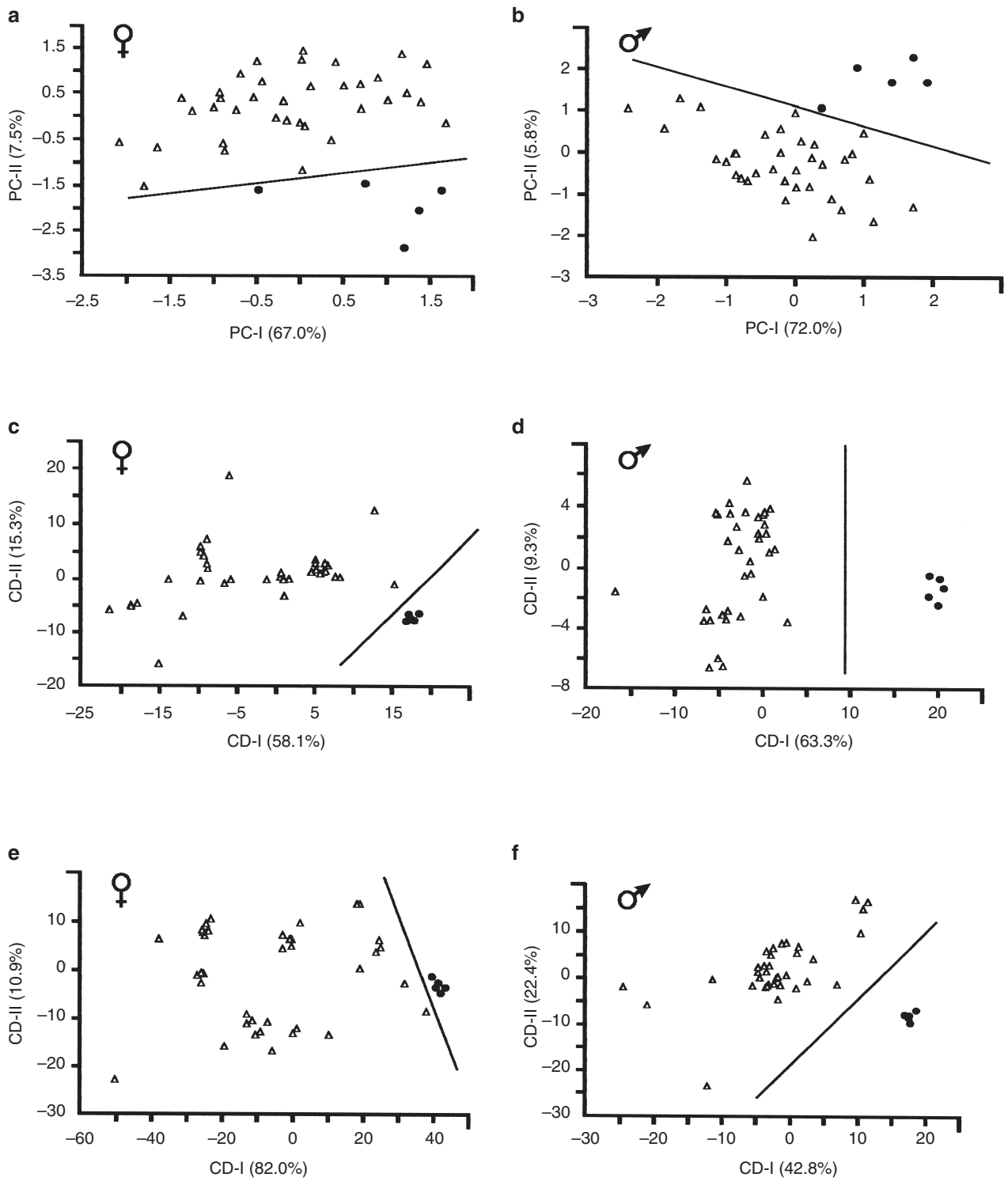

Fig. 2. Results of morphometric analysis. a–b, plots of first two principal components based on continuous data for *Encarsia* specimens (a, females; b, males). c–d, plots of first two canonical discriminant function based on continuous data for *Encarsia* specimens (c, females; d, males). e–f, plots of first two canonical discriminant function based on combined continuous and discrete data for *Encarsia* specimens (e, females; f, males). Δ *E. inaron*, • *E. estrellae*.

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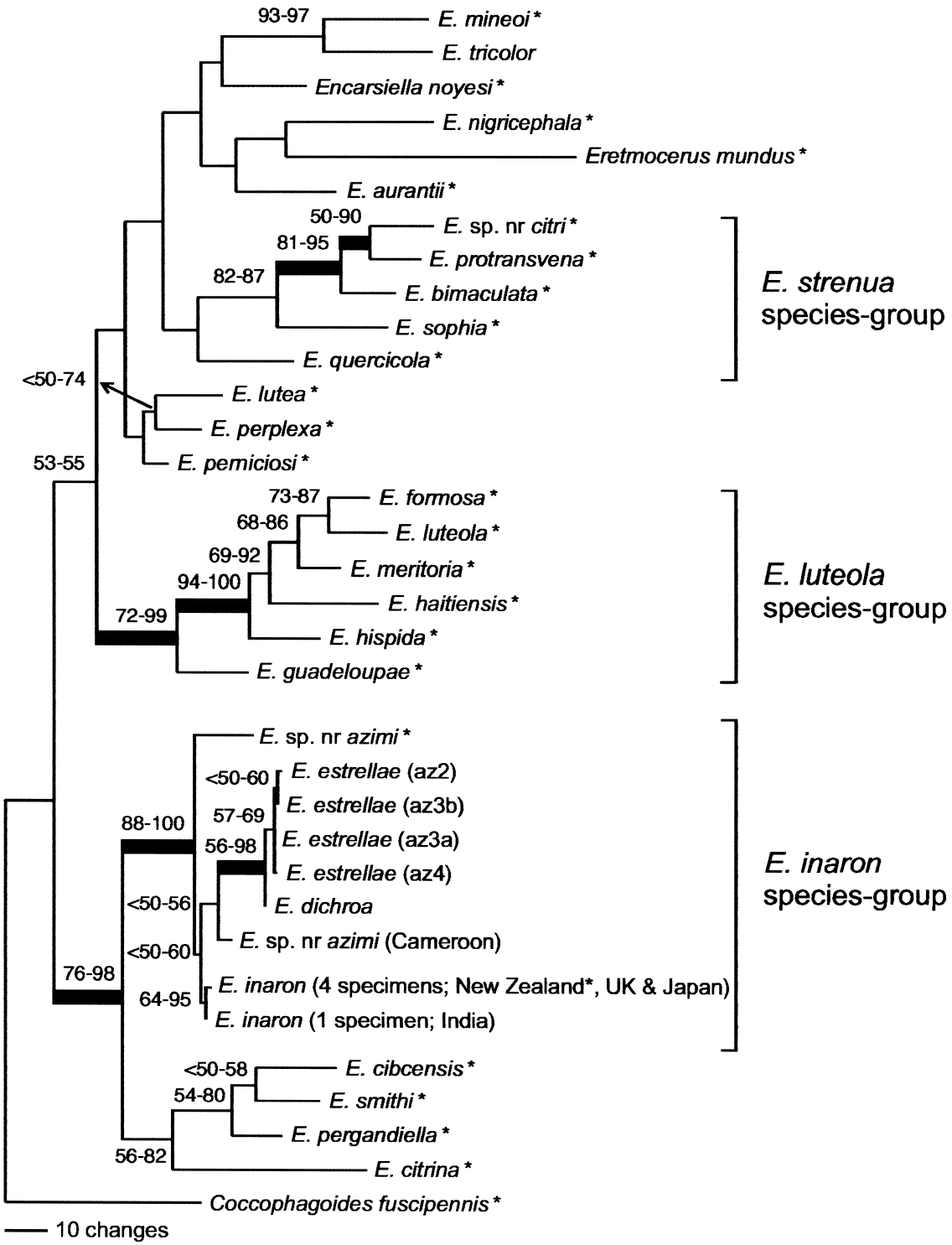

Fig. 3. One of 221 most parsimonious trees resulting from analyses of 28S-D2 rDNA sequence data (length = 866, R.I. = 0.641). Each sequence is from a single individual unless otherwise indicated. Branches are thickened when recovered in all MPTs from the five multiple alignments with gaps treated as both missing data and informative. Figures above branches show the range of bootstrap values (above 50%) for all analyses. An asterisk shows sequences from EMBL/GenBank databases (accession numbers: AF223366, AF223369–70, AF254192–94, AF254207, AF254211, AF254213, AF254221, AF254225–27, AF254230–32, AF254234–37, AF254242, AF254245–48, AF273667). See table 2 for accession numbers of new sequences (the inclusion of *Eretmocerus mundus* is here considerd to be an artefact).

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in fig. 3. Those branches that are also recovered in all MPTs are shown by thick lines. Within the *inaron* species-group, despite the existence of clear morphological differences between *E. dichroa* and *E. estrellae*, these are found to be more closely related than *E. estrellae* is to *E. inaron*. Our phylogenetic analysis confirms that *E. dichroa* and *E.* sp. near *azimi* (Cameroon) belong to the *E. inaron* species-group. Moreover, our sequence of *E.* sp. near *azimi* (Cameroon) differed from that of *E.* sp. nr *azimi* (Babcock *et al*., 2001) by 4.0–5.4% (depending upon different alignments; gaps treated as missing data), and from this it could be inferred that the specimens represent two different species.

Few sequence differences were found between individuals of *E. estrellae*: the average sequence distance based on different alignments was 0.54%, while the average sequence distance between *E. inaron* and *E. estrellae* was 3.6% (gaps treated as missing data).

The separation of *E. inaron* and *E. estrellae* was tested by constraining them in maximum parsimony analyses to form a clade (to the exclusion of *E. dichroa*). In this case, the shortest trees were between 6 and 20 steps longer than the unconstrained ones (gaps treated as informative), depending upon the alignment. In four of the five alignments, these constrained trees were significantly worse in Wilcoxon matched pairs sign tests implemented in PAUP* as the Templeton test ($P = 0.003–0.0596$ for all possible tree-to-tree comparisons; in the fifth alignment $P = 0.1573–0.2888$).

Within the genus *Encarsia*, overall relationships were similar to those found by Babcock *et al*. (2001), from whose study most of the sequences are derived. *Encarsia tricolor* was recovered with *E. mineoi* Viggiani in all except one of the most parsimonious trees, despite these two species having been placed in different species-groups (*E. tricolor* and *E. parvella* species-groups, respectively) (Heraty & Woolley, http://cache.ucr.edu/~heraty/Encarsia.cat.pdf). Although the genus *Encarsia* is never recovered as monophyletic, constraining it to be so resulted in most parsimonious trees that were significantly longer in only one of the five alignments (gaps treated as informative).

Discussion

Both the morphometric and molecular analysis indicate that the species from the Azores Islands is a new species, which is described below together with a redescription of *E. inaron* in the light of our analyses. The average sequence divergence within *E. estrellae* was typical of the values obtained by Babcock *et al*. (2001) for other *Encarsia* species, and the average sequence divergence between it and *E. inaron* was within the range of between-species values.

The first principal component in general is thought to be strongly influenced by overall size variation and possible allometric effects (Woolley & Browning, 1987). The host ranges for *E. inaron* and *E. estrellae* are variable (table 1), although some populations of *E. inaron* were restricted to a single host. Apart from the possible effect of hosts on general size, the discrimination of species was not affected by size (considering the separation based on other components). A canonical discriminant function appears to be the best means of separating these species. No single measurement or ratio provided a definite separation of the two species. *E. inaron* was most variable for both size and host range and the complexity of the separation is reflected by this variability.

The relative length of the marginal fringe, although playing an important role in *Encarsia* taxonomy, had almost no contribution in either sex to PC-I. Heraty & Polaszek (2000) also found that this character had little effect on PC-I for the *E. strenua* group.

Separation of E. estrellae *sp. n. and* E. inaron

Encarsia estrellae is morphologically very close to *E. inaron*, especially with respect to females. For females the main difference between the two species is the ratio of the third valvula length to the clava length. In *E. estrellae* the third valvula is less than 0.5 times as long as the clava, while in *E. inaron* it is at least 0.5 times as long as the clava. Male specimens can be distinguished by comparing the clava: the two claval segments are partially fused in *E. estrellae* (fig. 4c) but are separate in *E. inaron*. The main differences between the two species are summarized in table 4.

The *E. inaron* species-group, which was supported as a monophyletic group in this molecular analysis, has no defining suite of morphological characters. As already mentioned, this group has been characterized by having the ovipositor shorter than the mid tibia and basitarsus combined, a 2-segmented clava, and mid tibial spur less than 0.5 times basitarsus (Hayat, 1998). In this study, the latter character failed to place most *E. inaron* within the *E. inaron*-species group: in 59 female specimens, the mid tibial spur was on average 0.59 times as long as basitarsus. In addition, *E. dichroa*, which has a 3-segmented clava, was found to be a member of this group in the phylogenetic analysis.

All individual characters in DNA sequences, whether substitutions or insertion/deletion events, are prone to homoplasy, which means that single characters that define species-groups reliably are not available, however, two useful characters in the data can be emphasized. The following motif occurs 41 bases into the D2 region, and is found in all sequences of representatives of the *E. inaron* species-group (dashes represent gaps in the multiple alignment of all *Encarsia* species): AG(T/-)CCGCTTTG-GCTTCCGTGTGAA-CGCG. In positions 23–26 of this motif the AA-C was found only in representatives of the *E. inaron* species-group, and the insertion of a T in position 3 of this motif occurred only in *E. estrellae* and *E. dichroa* (no unambiguously aligned single character distinguished *E. estrellae* from all other *Encarsia* species).

Table 4. Summary of the main differences between *Encarsia estrellae* and *E. inaron*.

	E. estrellae	*E. inaron*
Female: third valvula length (TVL) / clava length (CLV)	TVL $< 0.5 \times$ CLV	TVL $\geqslant 0.5 \times$ CLV
Male: fifth and sixth flagellar segments (F5 and F6)	Partially fused (fig. 4c)	Separate

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Encarsia estrellae Manzari & Polaszek sp. n.

(fig. 4a–d)

Description. Female. Head dark yellow; occiput, areas behind postocellar bar brown; clypeus, malar space brown to dark brown. Mesosoma brown but pronotum, axillae and propodeum dark brown; middle of scutellum dark yellow to brown. Petiole brown, lateral parts dark brown. Gaster brown to dark brown. Third valvula yellow to dark yellow. Antenna dark yellow except scape, pedicel, anterior half of F5, F6 brown. Fore wings (fig. 4d) hyaline, slightly infuscate below marginal vein. Legs yellow except pretarsus brown. Antennal formula 1-1-4-2 (fig. 4b). Pedicel shorter than F1–F6 individually. Flagellum with the following numbers of longitudinal sensilla: F1:2, F2:3, F3:3, F4:4, F5:4, F6:3. Mid lobe of mesoscutum, axillae and scutellum with distinctly reticulate sculpture, longitudinal on the central scutellum (fig. 4a). Mid lobe of mesoscutum with 4+2+2 setae. Each side lobe of mesoscutum with 3 setae. Placoid sensilla on scutellum relatively distantly placed, distance between anterior pair of scutellar setae greater than that between posterior pair. Fore wing 2.41 times as long as wide (77:32). Marginal fringe of fore wing short. Tarsal formula 5-5-5. T1–T7 with 0+0, 1+1, 1+1, 1+1, 4, 4 and 5 setae, respectively. Ovipositor shorter than mid tibia and basitarsus combined (41:66), third valvula 0.36 times as long as second valvifer (11:30).

Male. Colour similar to female. Structural details essentially as for female, except ovipositor, and antenna with abundant longitudinal sensilla on all flagellomeres. The last two flagellar segments partially fused (fig. 4c). Male genitalia approximately as long as hind tibia.

Variation. In one female (ex *Bemisia afer*-group on *Hedera helix*), the gaster is largely pale, having only a single dark band on T6. Morphometrically this female and a (presumably conspecific) male agree with the remaining *E. estrellae* specimens, although no individual from this population was sequenced. Colour variation is well-documented in the closely-related *E. inaron* as a response to temperature experienced during development (Laudonia & Viggiani, 1993), and therefore could also be affected by altitude. These individuals are provisionally assigned to *E. estrellae*, pending further studies.

Species-group placement. *Encarsia inaron*-group.

Distribution. Azores Islands: Pico, São Miguel.

Hosts. Aleyrodidae: *Aleyrodes singularis* Danzig, *A.* ?*singularis*, *Bemisia* sp. *afer*-group. *Aleyrodes singularis* specimens from *Lysimachia nemorum* (Primulaceae) were positively identified by Dr J.H. Martin (BMNH). Specimens from the Azorean endemic *Euphorbia stygiana* (Euphorbiaceae) differ in some respects from *A. singularis*, and may represent a distinct, and possibly undescribed, species. In the north Atlantic islands the *Bemisia afer* group appears to be either highly diverse in terms of species, or highly morphologically variable within species. For this reason it has not been possible to identify to species-level some of the hosts of *E. estrellae*. The group is currently being studied in greater detail (J.H. Martin, BMNH, personal communication).

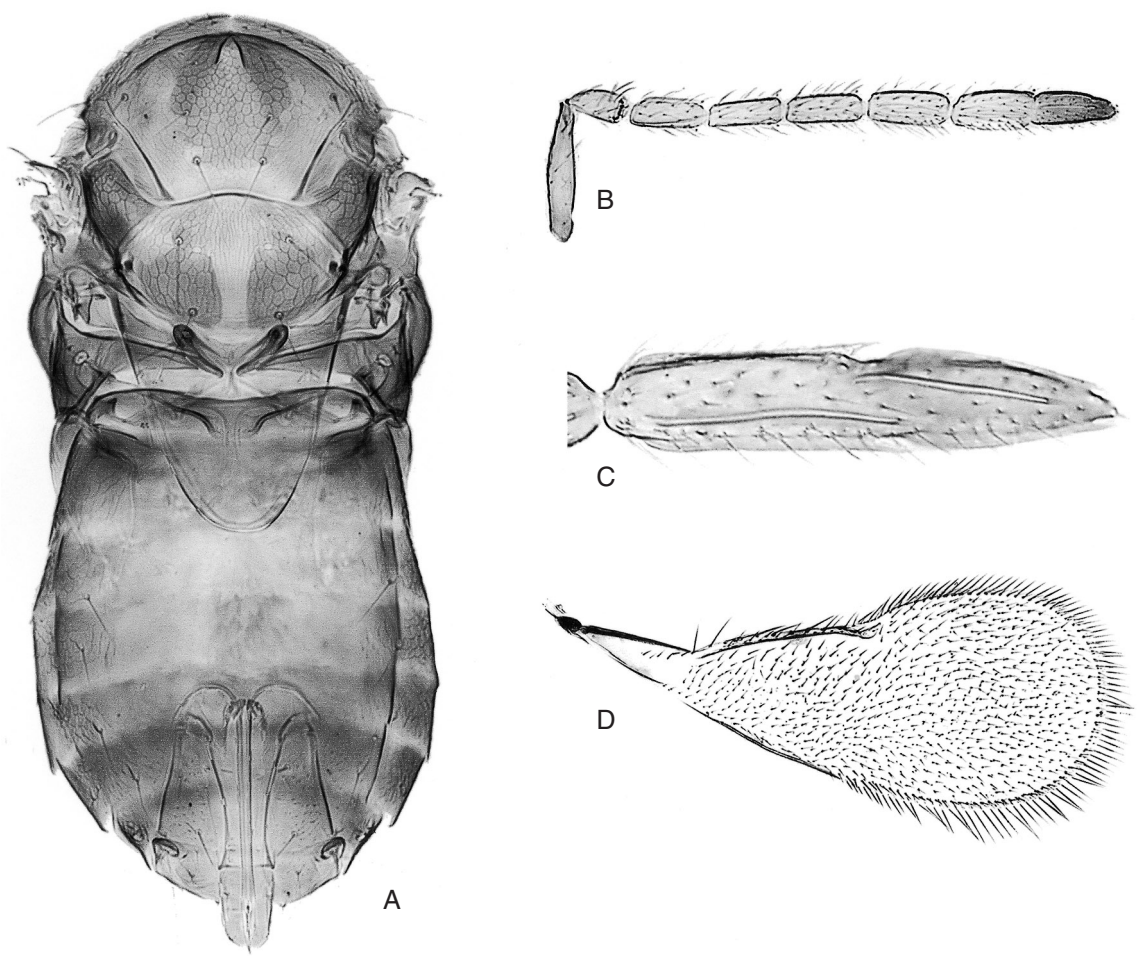

Fig. 4. *Encarsia estrellae* sp.n. A, mesosoma and gaster; B, antenna, female; C, F5+F6 of male antenna; D, fore wing.

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Material examined. Holotype ♀, AZORES, São Miguel, Lagoa Canarios, 715 m, 27.ix.1998, (E. Hernandez & A. Polaszek), ex *Aleyrodes singularis* on *Lysimachia nemorum* (BMNH). Paratypes 2♂, same data as holotype (BMNH). AZORES: São Miguel, Serra da Tronquiera, 2♀, 1♂, 26. ix.1998, (E. Hernandez & A. Polaszek) ex *Bemisia afer*-group on *Ilex perado azorica* (Aquifoliaceae) (BMNH). São Miguel, Sete Cidades, 1♀, 1♂, 27.ix.98, (E. Hernandez & A. Polaszek), ex *Bemisia* sp. on *Hedera helix canariensis* (Araliaceae) (BMNH). São Miguel, Serra da Tronquiera, 1♀, 1♂, 26.ix.1998, (E. Hernandez & A. Polaszek), ex *Aleyrodes singularis* on *Lysimachia nemorum* (BMNH). São Miguel, Serra da Tronquiera, 1♂, 26.ix.1998, (E. Hernandez & A. Polaszek), ex *Bemisia* sp. on *Viburnum tinus subcordatum* (Caprifoliaceae) (BMNH). Pico, Lagoa do Caiado 2♀, 6♂, 27.vi.00 (A. Polaszek) ex *Aleyrodes* ?*singularis* on *Euphorbia stygiana* (BMNH) (not used in morphometric analysis).

Encarsia inaron (Walker)

Description. Female. Head, mesosoma and petiole brown to dark brown. Gaster variable, from largely pale to largely brown. Third valvula pale. Antenna brown. Fore wing hyaline except bases. Legs yellow except coxae. In dark specimens mid and hind femora brown. Antennal formula 1-1-4-2. Pedicel shorter than F1–F6 individually. Flagellum with the following numbers of longitudinal sensilla: F1:0–4, F2:1–4, F3:1–4, F4:2–5, F5:2–5, F6:2–4. Mid lobe of mesoscutum, axillae and scutellum with distinctly reticulate sculpture, longitudinal on the central scutellum. Mid lobe of mesoscutum with 8–12 setae. Each side lobe of mesoscutum with three setae. Placoid sensilla on scutellum distantly placed, distance between anterior pair of scutellar setae greater than that between posterior pair. Fore wing 2.09–2.50 times as long as wide (90:43–17.5:7). Marginal fringe of fore wing short. Tarsal formula 5-5-5. T1–T7 with 0–3+0–4, 0–3+1–4, 0–4+0–4, 0–5+0–4, 4–11, 3–8 and 3–4 setae, respectively. Ovipositor shorter than mid tibia and basitarsus combined, third valvula 0.39–0.77 times as long as second valvifer (27:69–17:22).

Male. Entirely brown. Gaster sometimes pale. Structural details essentially as for female, except ovipositor, and antenna with abundant longitudinal sensilla on all segments. Antennomeres all separated. Male genitalia approximately as long as hind tibia.

Variation. See *E. estrellae* (above).

Species-group placement. *E. inaron*-group (= *E. partenopea*-group, *sensu* Viggiani & Mazzone, 1979).

Distribution. Europe, Africa, Asia, South America. Introduced into North America (Huang & Polaszek, 1998).

Hosts. Aleyrodidae: *Acaudaleyrodes rachipora* (Singh), *Aleurothrixus floccosus* (Maskell), *Aleyrodes lonicerae* Walker, *A. proletella* Linnaeus, *A. singularis*, *Asterobemisia carpini* (Koch), *A. paveli* (Zahradnik), *Bemisia tabaci* (Gennadius), *Bulgarialeurodes cotesii* (Maskell), *Pealius azaleae* (Baker & Moles), *P. quercus* (Signoret), *Siphoninus immaculatus* (Heeger), *S. phillyreae* (Haliday), *Trialeurodes vaporariorum* (Westwood).

Material examined. All specimens listed in table 1.

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References

Añez, N., Valenta, D.T., Cazorla, D., Quicke, D.J. & Feliciangeli, M.D. (1997) Multivariate analysis to discriminate species of phlebotomine sand flies (Diptera: Psychodidae): *Lutzomyia townsendi*, *L. spinicrassa*, and *L. youngi*. *Journal of Medical Entomology* **34**, 312–316.

Azidah, A.A., Fitton, M.G. & Quicke, D.L.J. (2000) Identification of the *Diadegma* species (Hymenoptera: Ichneumonidae, Campopleginae) attacking the diamondback moth, *Plutella xylostella* (Lepidoptera: Plutellidae). *Bulletin of Entomological Research* **90**, 375–389.

Babcock, C.S., Heraty J.M., De Barro, P.J., Driver, F. & Schmidt, S. (2001) Preliminary phylogeny of *Encarsia* Förster (Hymenoptera: Aphelinidae) based on morphology and 28S rDNA. *Molecular Phylogenetics and Evolution* **18**, 306–323.

Blackman, R.L. & Paterson, A.J.C. (1986) Separation of *Myzus* (*Nectarosiphon*) *antirrhinii* (Macchiati) from *Myzus* (*N.*) *persicae* (Sulzer) and related species in Europe (Homoptera: Aphididae). *Systematic Entomology* **11**, 267–276.

Blackman, R.L. & Spence, J.M. (1994) The effects of temperature on aphid morphology, using a multivariate approach. *European Journal of Entomology* **91**, 7–22.

Chishti, M.J.K. & Quicke, D.L.J. (1996) A revision of the Indo-Australian species of *Stenobracon* (Hymenoptera: Braconidae) parasitoids of lepidopterous stem borers of graminaceous crops. *Bulletin of Entomological Research* **86**, 227–245.

Evans, G.A. & Polaszek, A. (1998) The *Encarsia cubensis* species-group (Hymenoptera: Aphelinidae). *Proceedings of the Entomological Society of Washington* **100**, 222–233.

Evans, G.A., Polaszek, A. & Bennett, F.D. (1995) The taxonomy of the *Encarsia flavoscutellum* species-group (Hymenoptera: Aphelinidae) parasitoids of Hormaphididae (Homoptera: Aphidoidea). *Oriental Insects* **29**, 33–45.

Felsenstein, J. (1985) Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* **39**, 783–791.

Hayat, M. (1989) A revision of the species of *Encarsia* Foerster (Hymenoptera: Aphelinidae) from India and the adjacent countries. *Oriental Insects* **23**, 1–131.

Hayat, M. (1998) Aphelinidae of India (Hymenoptera: Chalcidoidea): a taxonomic revision. *Memoirs on Entomology, International* **13**, 416 pp.

Heraty, J.M. & Polaszek, A. (2000) Morphometric analysis and descriptions of selected species in the *Encarsia strenua* group (Hymenoptera: Aphelinidae). *Journal of Hymenoptera Research* **9**, 142–169.

Huang, J. & Polaszek, A. (1998) A revision of the Chinese species of *Encarsia* Förster (Hymenoptera: Aphelinidae): parasitoids of whiteflies, scale insects and aphids (Hemiptera: Aleyrodidae, Diaspididae, Aphidoidea). *Journal of Natural History* **32**, 1825–1966.

Laudonia, S. & Viggiani, G. (1993) Effetto della temperatura sulla colorazione degli adulti di *Encarsia partenopea* Masi (Hymenoptera: Aphelinidae). *Bollettino del Laboratorio di Entomologia Agraria 'Filippo-Silvestri' Portici*. (publ. 1995) **50**, 141–146.

Narusis, M.J. (1993) *SPSS for windows. Professional statistics*. 385 pp. SPSS Inc.

Polaszek, A. (1991) Egg parasitism in Aphelinidae (Hymenoptera: Chalcidoidea) with special reference to *Centrodora* and *Encarsia* species. *Bulletin of Entomological Research* **81**, 97–106.

https://doi.org/10.1079/BER2001144 Published online by Cambridge University Press

Polaszek, A., Evans, G.A. & Bennett, F.D. (1992) *Encarsia* parasitoids of *Bemisia tabaci* (Hymenoptera: Aphelinidae, Homoptera: Aleyrodidae): a preliminary guide to identification. *Bulletin of Entomological Research* **82**, 375–392.

Polaszek, A., Abd-Rabou, S. & Huang, J. (1999) The Egyptian species of *Encarsia* (Hymenoptera: Aphelinidae): a preliminary review. *Zoologische Mededelingen, Leiden* **73**, 131–163.

Quartau, J.A. (1982) A numerical analysis of character correlations in *Batracomorphus* Lewis (Insecta: Cicadellidae: Iassinae). *Boletim da Sociedade Portuguesa de Ciências Naturais* **21**, 51–58.

Swofford, D.L. (1998) *PAUP*. Phylogenetic analysis using parsimony (*and other methods)*, version 4. Sinauer Associates, Sunderland, Massachusetts, USA.

Viggiani, G. (1985) Additional notes and illustrations on some species of aphelinids described by A.A. Girault and A.P. Dodd in the genera *Coccophagus* Westw., *Encarsia* Foerst. and *Prospaltella* Ashm. (Hym.: Chalcidoidea). *Bollettino del Laboratorio di Entomologia Agraria 'Filippo Silvestri', Portici* **42**, 233–255.

Viggiani, G. (1987) Le specie italiane del genre *Encarsia* Foerster (Hymenoptera: Aphelinidae). *Bollettino del Laboratorio di Entomologia Agraria 'Filippo Silvestri', Portici* **44**, 121–179.

Viggiani, G. & Mazzone, P. (1979) Contributi alla conoscenza morfo-biologica delle specie del complesso *Encarsia* Foerster – *Prospaltella* Ashmead (Hym. Aphelinidae). *Bollettino del Laboratorio di Entomologia Agraria 'Filippo Silvestri', Portici* **36**, 42–50.

Woolley, J.B. & Browning, H.W. (1987) Morphometric analysis of uniparental *Aphytis* reared from chaff scale, *Parlatoria pergandii* Comstock, on Texas citrus (Hymenoptera: Aphelinidae; Homoptera: Diaspididae). *Proceedings of the Entomological Society of Washington* **89**, 77–94.

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