

1 **Title**

2 Evaluation of the effect of different insecticides on the survival and capacity of
3 Eretmocerus mundus Mercet to control Bemisia tabaci (Gennadius) populations
4

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23 **Abstract**

24 Two different experiments were carried out to evaluate three insecticides. In the
25 first one, the effect of two insecticides, methomyl and indoxacarb, on pupae and
26 adults of the whitefly Bemisia tabaci (Gennadius) parasitoid Eretmocerus mundus
27 Mercet was evaluated under laboratory and greenhouse conditions, using sweet
28 pepper (Capsicum annuum L.) plants. In the second experiment, oxamyl was
29 tested to study its effect on the ability of E. mundus to parasitize and control B.
30 tabaci in sweet pepper plants, using a greenhouse cage evaluation. Methomyl and
31 indoxacarb caused low mortality of E. mundus pupae (17.6 and 7.8%
32 respectively), although methomyl mortality was significantly higher. Methomyl
33 produced 100% mortality on E. mundus adults with fresh and 24 hour-old
34 residues on leaves, significantly higher than the mortality produced by indoxacarb
35 (values ranged from 43.9 to 34.4%). The harmful effect of methomyl persisted for
36 a long time (up to 60 days). The results of the experiment with oxamyl showed
37 that E. mundus controlled whitefly population, without significant interaction
38 between the presence of the parasitoid and insecticide on whitefly mortality.
39 Whitefly mortality in the presence of the parasitoid was 87.8%, significantly higher
40 than the mortality in the absence of E. mundus (59.3%). Oxamyl did not produce
41 a significant effect on the emergence of E. mundus adults. Application of the
42 products in IPM programs is discussed.

43

44 **Keywords:** Eretmocerus mundus, Bemisia tabaci, indoxacarb, oxamyl, methomyl, sweet
45 pepper, parasitism.

46

47 **Introduction**

48

49 Eretmocerus mundus Mercet (Hymenoptera: Aphelinidae), a parasitoid of the
50 tobacco, cotton or sweet potato whitefly Bemisia tabaci (Gennadius) (Hemiptera:
51 Aleyrodidae), has a widespread distribution worldwide. This species is recognized
52 as one of the most important natural enemies of B. tabaci, and has generated a
53 lot of interest in countries where B. tabaci is a problem. The use of heavy chemical
54 control against this pest is also recognized to be involved in many of the outbreaks
55 of B. tabaci around the world, for different reasons: development of insecticide
56 resistance, negative effects on natural enemies, alteration of behaviour and
57 biology of the pest.

58

59 Outbreaks of this whitefly in different crops and countries have stimulated the
60 search for different biological control agents around the world. The most active
61 country in this respect is the U.S.A., where many parasitoid and predator species
62 have been tested. One of the most promising species is E. mundus (Goolsby et al.,
63 1998; Hoelmer et al., 1999; Kirk et al., 2000), especially the strain obtained from
64 southeast Spain. The first time this species was collected in Murcia (Spain) on
65 cotton, in 1991-92, it exhibited a good tolerance to the different pesticides applied
66 regularly to the crop (Kirk et al., 2000).

67

68 Current pest control includes the use of pesticides, combined or not with natural
69 enemies. There are many reports and studies that present evidence of natural
70 enemies of B. tabaci that can remain active in different crops after the use of
71 some pesticides (Gerling, 1996; Gerling and Naranjo, 1998; Simmons and Jackson,
72 2000), especially parasitoids, such as E. mundus. Possible explanations for this
73 ability of parasitoids to remain in treated plots include the protection that
74 immature stages can obtain inside the host, as well as the type of compound
75 used, the incomplete coverage of the canopy or the timing of the application.

76

77 The presence of E. mundus in commercial plots treated routinely with pesticides
78 has also been reported by other authors (Rodriguez-Rodriguez et al. 1994;

79 Gonzalez-Zamora et al., 1996) in southeast Spain. They also studied the
80 parasitism on B. tabaci and Trialeurodes vaporariorum (West.) by different species
81 and the ability of E. mundus to control B. tabaci populations in sweet pepper,
82 melon, and tomato. In summary, these papers show the ability of different
83 whitefly parasitoids to establish and maintain a high level of parasitism in plots,
84 even when they are treated with pesticides. Different pesticides are considered to
85 be selective to this and other parasitoids and can be used together to obtain
86 control of the pest population below injury levels (Koppert Biological Systems,
87 2003).

88

89 Other papers have studied the combination of pesticides and parasitoids (with
90 special interest in Eretmocerus spp.) to control whitefly populations in different
91 conditions. For example, Birnie and Denholm (1992) used an extended laboratory
92 trial to test the capacity of E. mundus to control B. tabaci populations on cotton
93 with the application of cypermethrin, showing the ability of the parasitoid
94 population to recover after only one application of the compound. Devine et al.
95 (2000) revealed the potential of piperonyl butoxide to improve the level of E.
96 mundus parasitism on B. tabaci, by slowing the development of the whitefly,
97 increasing the parasitism in the treated whitefly population by 7-8%. Van Driesche
98 et al. (2001) demonstrated the possibility of using an insect growth regulator
99 (buprofezin) in combination with Eretmocerus eremicus Rose and Zolnerowich to
100 control T. vaporariorum and B. tabaci in poinsettias in commercial greenhouses.

101

102 The aim of this work was to study the effect of three insecticides on different
103 development stages (pupa and adult) of E. mundus, and the capacity of E.
104 mundus to control B. tabaci populations together with one of the insecticides,
105 presenting evidence of the capacity of this species to be used together with
106 insecticides. Two of these insecticides (methomyl and oxamyl) are currently used
107 in southeast Spain to control different pests, and the third (indoxacarb) has been
108 introduced recently to control caterpillars in different crops, including those with B.
109 tabaci infestation. Indoxacarb is a selective product that is compared with the
110 other two products, non-selective insecticides. These two insecticides can be

111 applied in different ways in order to confer selectivity. Results are discussed
112 considering the possibilities of using these products in a IPM program.

113

114 **Material and Methods**

115

116 Two different experiments were carried out to test the insecticides: in the first
117 one, methomyl (chemical name: S-methyl-N-[(methylcarbamoyl)oxy]
118 thioacetimidate) (commercial product Lannate[®] 20 L, Du Pont Iberica S.L.), and
119 indoxacarb (chemical name: (S)-7-chloro-3-[methoxycarbonyl-(4-trifluoromethoxy-
120 phenyl)-carbamoyl]-2,5-dihydro-indenol 1,2 -e[1,3,4]oxadiazine-4a(3H)-carboxylic
121 acid methyl ester) (commercial product Steward[®] 30 WG, Du Pont Iberica S.L.)
122 were tested on E. mundus pupae and adults; in the second one, oxamyl (chemical
123 name: methyl-N',N'-dimethyl-N[(methylcarbamoyl)oxy]-1-thio-oxamimidate)
124 (commercial product Vydate[®] 10L, Du Pont Iberica S.L.) was tested to determine
125 its effect on the parasitism and capacity of E. mundus to control a population of
126 B. tabaci, using a greenhouse cage evaluation.

127

128 A colony of B. tabaci was created from adults captured in the Seville province
129 (southwest Spain), from cotton and aubergine plants, and kept in an insect
130 rearing room, feeding on sweet pepper (Capsicum annuum L.) plants (cv. Largo
131 Italiano, Semillas Batlle S.A., C/Matadero 10, E-10100-Miajadas (Caceres, Spain)).
132 The biotype of B. tabaci was not determined. Adults of E. mundus were collected
133 in the regions of Almeria (southeast Spain) and Seville, on sweet pepper and
134 cotton plants, and reared on sweet pepper plants infested with B. tabaci nymphs.
135 The rearing room was kept at 26±2 °C with a photoperiod of 16:8 (Light:Dark).

136

137 **Experiment 1: Effect of methomyl and indoxacarb on Eretmocerus** 138 **mundus pupae and adults**

139

140 Four young sweet pepper plants were selected for each treatment (i.e. product),
141 with 4-5 leaves each, to assess the effect of insecticides on E. mundus pupae.
142 They were infested with adults of B. tabaci, which were allowed to lay eggs for

143 48-72 hours and then removed from the plants. Development of the offspring was
144 followed until 2nd-3rd instar nymphs were present, then females of E. mundus
145 were introduced into the cages for oviposition, together with several males. The
146 plants were kept in the rearing room until the parasitoid pupa was observed inside
147 the whitefly pupal case: the form of the pupa was evident and the eyes took on a
148 cherry colour (Garrido et al., 1982; Garrido, 1992). At that moment, a minimum of
149 twenty-five pupae per plant were marked with an indelible pen, and the plants
150 were put in a plastic greenhouse. The total number of pupae used in each
151 treatment was 115, 107, and 114 for indoxacarb, methomyl and the control,
152 respectively. The products were applied in that moment, using a trigger-operated
153 hand sprayer, at the recommended field rates: methomyl, 0.4 g a.i./l; indoxacarb,
154 0.0375 g a.i./l until run-off. A group of four plants was treated with water, as
155 control.

156

157 Counts were done every 3-4 days with the help of a 5x magnifying hand lens,
158 counting dead and live pupae and pupal cases from where an insect had emerged.
159 The experiment finished when all the adults had emerged.

160

161 The effect of the insecticides on E. mundus adults was evaluated by applying the
162 products and water at the same rates with the trigger-operated hand sprayer on
163 several sweet pepper plants with developed leaves. The products were allowed to
164 dry and then two types of cages were mounted: with fresh residues and with
165 twenty-four hour old residues. The methodology used was described in Jones et
166 al. (1995).

167

168 The experimental units (cages) consisted of 5-6 newly emerged adults of E.
169 mundus, confined to a Petri dish of 55 mm diameter and 14 mm height. The Petri
170 dishes were modified by replacing most of the bottom with organdy cloth
171 (subsequently inverted to become the top). The new bottom half of the Petri dish
172 was used as a template to cut out a circular section of sprayed leaf. The leaf discs
173 were fitted in the inner surface of a dish lid (now the bottom), with the bottom
174 leaf surface facing upwards. A cotton cloth moistened with water and honey was

175 put inside the cage. The parasitoids were collected from the rearing units and
176 briefly chilled before being introduced in the experimental units. The ventilated
177 dish bottom was replaced, becoming the top. Dish halves were secured using a
178 pair of clips, and the cages were put in a laboratory room. Six replicates per
179 treatment (with a total of 32-36 individuals per treatment) were used with the
180 fresh residues leaves, and eight replicates per treatment (with a total of 53-61
181 individuals per treatment) with the leaves with twenty-four hour old residues. The
182 adults were kept in the cages for twenty-four hours, then opened and the adults
183 counted, separating dead and live insects.

184

185 A similar test was performed to study methomyl persistence in sweet pepper plant
186 leaves. Different groups of plants were selected, and in each group one of the
187 plants was treated with water and the others with methomyl at the previous rate.
188 The plants were allowed to dry and after different time periods, ranging from
189 seven to sixty-two days after treatment, cages were mounted with leaf discs from
190 plants treated with methomyl and water. The number of adults introduced in each
191 cage varied between eight and thirteen. The adults were introduced in the cages
192 as explained above, and after twenty-four hours the cages were opened and the
193 number of dead and live adults counted. Treated plants and cages were kept in
194 laboratory conditions.

195

196 **Experiment 2: Effect of oxamyl on the capacity of Eretmocerus mundus**
197 **to control *Bemisia tabaci***

198

199 The experimental design studied two levels (presence and absence) of two factors
200 (parasitoid and insecticide). There were, therefore, four treatments: 1) application
201 of oxamyl, 2) application of oxamyl and introduction of E. mundus, 3) introduction
202 of E. mundus, and 4) no application of oxamyl and no introduction of E. mundus
203 (control). Each treatment was replicated four times, using one sweet pepper plant
204 per treatment and replicate, making a total of 16 plants.

205

206 The plants were infested with B. tabaci adults in the rearing room, allowed to lay
207 eggs for three days only on one leaf and then removed. The plants were kept in
208 the rearing room and, after nine days, first and second instar whitefly nymphs
209 were present on the leaves. The number of nymphs per leaf ranged from 132 to
210 198. Plants were then maintained in a greenhouse inside a metallic structure
211 covered with organdy for the rest of the experiment.

212

213 Oxamyl was applied at a rate of 0.028 g a.i./plant (equivalent to 500 g a.i./ha,
214 with 18,000 plants/ha) in treatments 1) and 2). It was applied on a weekly basis,
215 a maximum of eight times, beginning the same day the plants had been put in the
216 greenhouse. The oxamyl was diluted in 100 ml of water per plant. The pH of the
217 broth was adjusted to 4.5-5.5 with phosphoric acid. A drip irrigation system was
218 used to water the plants, simulating the normal irrigation management of this
219 crop, and the broth was injected with a previous-pressure sprayer in the irrigation
220 system.

221

222 E. mundus was introduced in treatments 2) and 3) at a total rate of 12 to 18
223 females per replicate, following the recommended ratio proposed by Jones et al.
224 (1999). Adult parasitoids were placed in the cages in two to four separate
225 introductions, beginning on the first day that the plants were put in the
226 greenhouse.

227

228 Plants were evaluated every 3-4 days during the first two weeks, and then weekly
229 until all the adults of B. tabaci and E. mundus emerged. The number of living
230 whitefly nymphs and pupae, the number of parasitized whitefly nymphs and
231 parasitoid pupae, and the number of pupal cases from where an adult (whitefly or
232 parasitoid) had emerged were counted. This experiment included only one
233 generation of the whitefly B. tabaci and the parasitoid E. mundus.

234

235 Analysis of variance was performed on mortality and parasitism (Statistical
236 Graphics Corporation, 1999) in both experiments, with the transformation of
237 $z = \arcsin \sqrt{p}$, where p is mortality or parasitism. A 2x2 factorial analysis was applied

238 in experiment 2. If treatments were significant at $P < 0.05$, then differences
239 between means were determined using the LSD test at 95% confidence level.
240 Abbot's formula (Abbot, 1925) was used to correct the mortality in experiment 1.

241

242 Voucher specimens of the parasitoid are maintained by the first author in the
243 Department collection (Departamento de Ciencias Agroforestales, Universidad de
244 Sevilla).

245

246 **Results**

247

248 **Experiment 1: Effect of methomyl and indoxacarb on Eretmocerus** 249 **mundus pupae and adults**

250

251 Methomyl significantly increased the mortality of E. mundus pupae parasitizing B.
252 tabaci (Figure 1) seven days after application ($F=4.7$, d.f.=2, 9, $P=0.04$), while
253 indoxacarb was not significantly different from the control. Mortality increased
254 three days later to 17.6 % in methomyl, greater than indoxacarb and the control,
255 with 7.8 and 5.1% mortality respectively ($F=8.20$, d.f.=2, 9, $P=0.009$).

256

257 Both insecticides significantly increased the mortality of E. mundus adults (Figure
258 2), though methomyl was more harmful (100 % mortality in both cases, with fresh
259 and 24 hour-old residues in leaves) than indoxacarb (43.9% and 34.4%,
260 respectively), with $F=59.8$, d.f.=2, 6, $P<0.0001$, and $F=84.60$, d.f.=2, 9,
261 $P<0.0001$, for fresh and 24 hour-old residues respectively.

262

263 The harmful effect of methomyl on E. mundus adults lasted up to sixty days after
264 application (Figure 3A), when the mortality was still significantly higher than the
265 control treated with water. The mortality was corrected with the Abbot's formula
266 and adjusted to a curve (Figure 3B; $R^2 = 0.88$; $P<0.01$). At least 55 days were
267 required for adult mortality after treatment with methomyl to drop to 50%.

268

269 **Experiment 2: Effect of oxamyl on the capacity of Eretmocerus mundus**
270 **to control Bemisia tabaci**

271

272 The percentage mortality of B. tabaci nymphs treated with oxamyl was not
273 significantly different from untreated nymphs ($F=0.05$, d.f.=1, 12, $P=0.83$). The
274 addition of E. mundus produced a significant effect on the mortality of B. tabaci
275 nymphs ($F=6.90$, d.f.=1, 12, $P=0.02$). No interaction was observed between the
276 two factors studied ($F=2.69$, d.f.=1, 12, $P=0.13$).

277

278 The results of whitefly mortality in the four treatments are shown in Figure 4A.
279 The highest mortality was obtained with the introduction of E. mundus without
280 application of oxamyl (95.7% at the end of the study), whereas the mortality
281 decreased to 79.9% when the plants were treated with the insecticide. The
282 natural mortality of the whiteflies was 51.3% and the mortality in the treatment
283 with oxamyl reached 67.3%. There is no significant difference between the four
284 treatments ($F=3.21$, d.f.=3, 12, $P=0.06$), but the P-value is very near to the limit
285 of $P=0.05$. Although not completely justified, Tukey's HSD test only showed
286 differences between treatment 3 (introduction of E. mundus, 95.7% mortality) and
287 treatment 4 (control, 51.3% mortality).

288

289 Levels of whitefly mortality in the treatments with and without E. mundus are
290 shown in Figure 4B. The values at the end of the study were 87.8 and 59.3%
291 respectively, which differ significantly ($F=6.90$, d.f.=1, 12, $P=0.02$).

292

293 The number of fourth instar nymphs with signs of parasitization was also recorded
294 in the two treatments where E. mundus was used (Figure 5A). This was very
295 similar in the treatments with and without oxamyl, although without oxamyl the
296 proportion of parasitized nymphs was always higher. In any case, the standard
297 error bars superimpose in most of the counting dates, indicating that there was no
298 significant difference between the two treatments.

299

300 Finally, the proportion of parasitoids that emerged at the end of the experiment
301 from whitefly pupal cases was higher in plants not treated with oxamyl (Figure
302 5B), but not significantly different from treated plants (24.7% and 10.6%,
303 respectively; $F=4.11$, $d.f.=1, 6$, $P=0.09$). The final value was measured in terms
304 of the proportion of the initial nymphal population.

305

306 **Discussion**

307

308 The results reveal that indoxacarb did not significantly increase E. mundus pupal
309 mortality, while methomyl presented a low mortality. A similar pattern has been
310 found with different insecticides tested on E. mundus pupae. Gonzalez-Zamora et
311 al. (1997) found that only three of thirteen insecticides tested were included in the
312 category of moderately harmful (between 80 and 98% mortality, Abbott's
313 corrected mortality), according to the I.O.B.C. classification. On the other hand,
314 Jones et al. (1998), testing six insecticides on E. mundus pupae, found that three
315 of them produced a mortality higher than 90 %, and the other three of around or
316 below 50%. The fact that the insect develops inside the pupal case of the whitefly
317 can help to explain the low mortality produced by some of the insecticides tested.
318 The timing of application of the compounds, which is related to the development
319 stage of the parasitoid, and their ways of action can also help to explain the
320 different effect on the parasitoid, as discussed by Gerling and Sinai (1994) and
321 Jones et al. (1998). Gerling and Sinai (1994) observed that buprofezin was toxic to
322 the eggs and young larvae of Eretmocerus sp., but harmless to the parasitoid
323 pupae, and Jones et al. (1998) also found differential mortalities of the products
324 they tested depending on the stage of the parasitoid, as young larva or parasitoid
325 pupa. In the present work, the compounds were applied when pupae were easily
326 observed, and all the individuals were in the same stage, in order to have an
327 homogeneous population.

328

329 This result, however, cannot be generalized for all parasitoids and products.
330 Garrido et al. (1982) found high mortality (97.2%) on Cales noacki Howard pupae
331 (Hymenoptera: Aphelinidae) parasitizing Aleurothrixus floccosus Maskell

332 (Hemiptera: Aleyrodidae) treated with methomyl. In other parasitoids such as
333 Aphidius colemani Vierek (Hymenoptera: Aphidiidae) parasitizing Myzus persicae
334 (Sulzer) (Hemiptera: Aphididae), methomyl showed no effect on the emergence of
335 adults from inside the mummies (DuPont, interim report, 1999). Indoxacarb also
336 showed the same low toxicity with other parasitoids, such as aphid parasitoids
337 developing inside the mummy (Dinter and Wiles, 2000).

338

339 The two compounds tested with E. mundus adults showed a different pattern
340 (Figure 2). Indoxacarb produced a low mortality with fresh and 24 hour-old
341 residues (43.9 and 34.4% respectively), whereas methomyl produced 100%
342 mortality in both cases. Indoxacarb has been tested on different beneficial
343 arthropods (Dinter and Wiles, 2000), and proved harmless to Typhlodromus pyri
344 Scheuten (Acari: Phytoseiidae), Episyrphus balteatus De Geer (Diptera:
345 Syrphidae), Orius laevigatus (Fieber) (Hemiptera: Anthocoridae) and Aleochara
346 bilineata Gyllenhal (Coleoptera: Staphylinidae), but harmful to parasitic wasps (A.
347 colemani) under worst-case laboratory conditions (the same conditions as in the
348 present study). In extended and semi-field trials it proved to be compatible with
349 the presence of different aphid parasitoids. The conclusion of the authors was that
350 this product could be included in integrated pest management programs. Our
351 results indicate a low impact of this product on both E. mundus adult and pupa.

352

353 On the other hand, our results reveal that methomyl is a very toxic compound for
354 E. mundus adults, although this result may be different with other insects.
355 Methomyl produced high mortality in A. colemani adults, under worst-case
356 laboratory conditions after 24 hour exposure to fresh residues, although this effect
357 lasted around two weeks when the mortality dropped to 25-50% (DuPont, interim
358 report, 1999). In laboratory and field conditions, methomyl showed partial and
359 short-term effects on foliage- and surface-dwelling predators, such as larvae of
360 Chrysoperla carnea (Stephens) and Coccinella septempunctata L., in which the
361 mortality was comparable to the control 4 days after the treatment (Dinter and
362 Kratz, 2000).

363

364 The harmful effect of methomyl residues on adults of E. mundus clearly shows a
365 long lasting negative effect on this parasitoid (Figure 3). About 55 days were
366 necessary for adult mortality to decline below 50%. This means that adults
367 emerging from pupae onto treated foliage will not survive, considering that the
368 time spent on pupa is about 6 to 15 days at 25 °C (Foltyn and Gerlin, 1985;
369 Sharaf and Batta, 1985). On the other hand, it is necessary to consider the effects
370 of light intensity and crop growth on product degradation and dilution of the
371 residues. In commercial crops, the time to reach the 50% mortality could be
372 shorter for these reasons and, also, parasitoids may not come into contact with
373 treated foliage, simply because their hosts are present on younger growth, or
374 leaves treated early in the crop have senesced or been removed. Anyway, 55-56
375 days (or 8 weeks) is the minimum time some companies recommend to wait
376 before introducing parasitoids such as E. eremicus (and other species) after a
377 treatment with methomyl (Koppert Biological Systems, 2003).

378
379 The second experiment, with oxamyl, showed interesting results: (1) the control
380 that E. mundus can exert on a B. tabaci population, and (2) the absence of a
381 significant interaction between the parasitoid and the insecticide.

382
383 In this trial, the presence of E. mundus produced a mortality of whitefly nymphs of
384 87.8%, significantly different from the absence of the parasitoid. The whitefly
385 mortality data alone cannot tell us the fate of the population, because experiment
386 2 only included one generation of the whitefly and parasitoid. Different authors
387 (Birnie and Denholm, 1992; Simmons and Minkenberg, 1994; Goolsby et al., 1998;
388 Heinz and Parrella, 1998) have shown the ability of E. mundus, and other related
389 species, to control whitefly populations in extended laboratory and semi-field
390 experiments. These authors kept whiteflies in cages and released adult
391 parasitoids, albeit in different proportions than in the present work, and generally
392 for more than one generation.

393
394 The high whitefly mortality when the parasitoid is present is caused by active
395 parasitism and the feeding activity of the adult parasitoids. This last component

396 can be very important, as Heinz and Parrella (1998) showed for different adult
397 parasitoids, including E. mundus. The final result is that the whitefly population
398 reaches low numbers, significantly lower than the control in which no parasitoid is
399 added.

400

401 Oxamyl exerted little control on the B. tabaci population, compared with the
402 untreated control in the conditions of the experiment, but it must be noted that
403 nymphs were already installed on the leaves when the product was applied.
404 Oxamyl is a product that can control whitefly populations (and other sucking
405 pests) on a long-term basis, as Cabello et al. (1997) demonstrated. These authors
406 found significant differences between treated and untreated plots after 56 days
407 from the beginning of the treatments, whereas in our case we only studied one
408 generation of the population of B. tabaci and E. mundus.

409

410 The absence of a significant interaction between the two factors (parasitoid and
411 insecticide), measured in terms of whitefly mortality, was also observed. The
412 combination of oxamyl and a whitefly parasitoid was also studied by Helyer et al.
413 (1984) using Encarsia formosa Gahan to control T. vaporariorum in tomato,
414 finding that the level of parasitism was 90% 145 days after treatment with
415 oxamyl, although they only applied the product once and only introduced the
416 parasitized pupae of E. formosa after 56 days. In the present study, oxamyl did
417 not produce a significant effect on the proportion of final emergence of E. mundus
418 adults, although the P value obtained (0.09) was near to the limit of $P= 0.05$. The
419 evolution of parasitized nymphs over the sampling dates was rather similar in the
420 treated and untreated plots.

421

422 Finally, our results suggest that indoxacarb could be used in integrated pest
423 management programs for crops where E. mundus is present. Oxamyl could also
424 be considered, although the effect on the parasitoid population in the longer term
425 should be studied. Methomyl is very toxic to E. mundus adults, and the use of this
426 product in crops where this parasitoid is an important agent to control B. tabaci
427 should be avoided, especially when whitefly is a key pest. In other cases, aspects

428 such as the crop, the type of pest or pests to control, the timing of pesticide
429 application and location on the plant, or the relation between pests and natural
430 enemies present in the crop should be studied to use or reject this product.

431

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433

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436

Final Version

437 **References**

438

439 Abbot, W.S. 1925. A method of computing the effectiveness of an insecticide. J.
440 Econ. Entomol. 18, 265-267.

441

442 Birnie, L. C. and Denholm, I. 1992. Field simulators: a novel approach to
443 evaluating the impact of pesticides on beneficial arthropods in the laboratory.
444 Aspects of Applied Biology 31, 105-112.

445

446 Cabello Garcia, T., Gomez, M., Barranco, P., Lucas, M., and Belda, J. E. 1997.
447 Evaluation of oxamyl against homoptera pests in greenhouse-grown pepper,
448 applied with drip irrigation. Ann. appl. Biol. 130 (supplement) Test of
449 Agrochemicals and Cultivars 18, 2-3.

450

451 Devine, G. J., Wright, D. J., and Denholm, I. 2000. A parasitic wasp (Eretmocerus
452 mundus Mercet) can exploit chemically induced delays in the development rates of
453 its whitefly host (Bemisia tabaci Genn.). Biol. Control 19, 64-75.

454

455 Dinter, A., and Kratz, T. 2000. Impact of field treatments with the insecticide
456 Lannate 20 L (a.i. methomyl) on foliage and ground dwelling arthropods.
457 IOBC/WPRS Bull. 23, 127-135.

458

459 Dinter, A., and Wiles, J.A. 2000. Safety of the new DuPont insecticide
460 "Indoxacarb" to beneficial arthropods: an overview. IOBC/WPRS Bull. 23, 149-156.

461

462 Foltyn, S., and Gerling D. 1985. The parasitoids of the aleyrodid Bemisia tabaci in
463 Israel: development, host preference and discrimination of the aphelinid wasp
464 Eretmocerus mundus. Entomol. exp. appl. 38, 255-260.

465

466 Garrido, A. 1992. Pesticides toxicity over pupae of Cales noacki Howard (Hym.:
467 Aphelinidae). IOBC/WPRS Bull. 15, 56-60.

468

469 Garrido, A., Tarancon, J., and del Busto, T. 1982. Incidencia de algunos
470 plaguicidas sobre estados ninfales de Cales noacki How., parasito de Aleurothrixus
471 floccosus Mask. An. INIA-Ser. Agric. 18, 73-96.
472

473 Gerling, D. 1996. Status of Bemisia tabaci in the Mediterranean countries:
474 opportunities for biological control. Biol. Control 6, 11-22.
475

476 Gerling, D., and Naranjo, S. E. 1998. The effects of insecticide treatments in
477 cotton fields on the levels of parasitism of Bemisia tabaci (Gennadius) s.l. Biol.
478 Control 12, 33-41.
479

480 Gerling, D., and Sinai, P. 1994. Buprofezin effects on two parasitoid species of
481 whitefly (Homoptera: Aleyrodidae). J. Econ. Entomol. 87, 842-846.
482

483 Gonzalez-Zamora, J. E., Gallardo, J. M., and Garcia, M. M. 1997. Toxicity of
484 different pesticides on pupae of Eretmocerus mundus Mercet (Hymenoptera:
485 Aphelinidae) parasitizing Bemisia tabaci (Genn.) (Homoptera: Aleyrodidae).
486 IOBC/WPRS Bull. 20, 114-120.
487

488 Gonzalez-Zamora, J.E., Rodriguez-Rodriguez, M. D., Mirasol Carmona, E.,
489 Manzanares Ruiz, C., Moreno Vazquez, R., Rodriguez Rodríguez, M. P., and
490 Lastres Garcia-Teston, J. 1996. Evolución del parasitismo en Bemisia tabaci
491 (Genn.) y Trialeurodes vaporariorum (West.) (Homoptera: Aleyrodidae) en
492 invernaderos en Almería. Bol. San. Vegetal-Plagas 22, 373-389.
493

494 Goolsby, J., Ciomperlink, M. A., Legaspi, B. C., Legaspi, J. C., and Wendel, L. E.
495 1998. Laboratory and field evaluation of exotic parasitoids of Bemisia tabaci
496 (Gennadius) (Biotype "B") (Homoptera: Aleyrodidae) in the Lower Rio Grande
497 Valley of Texas. Biol. Control 12, 127-135.
498

499 Heinz, K. M., and Parrella, M. P. 1998. Host location and utilization by selected
500 parasitoids of Bemisia argentifolii (Homoptera: Aleyrodidae): implications for
501 augmentative biological control. Environ. Entomol. 27, 773-784.
502

503 Helyer, N. L., Ledieu, M. S., Hussey, N. W., and Scopes, N. E. A. 1984. Early
504 season integrated control of whitefly on tomatoes using oxamyl. British Crop
505 Protection Conference Monograph, 293-297.
506

507 Hoelmer, K., Kirk, A., and Simmons, G. 1999. An overview of natural enemy
508 explorations and evaluations for Bemisia in the U.S. Proceedings of the Fifth
509 International Conference on Pests in Agriculture-ANPP, 7-9 December 1999,
510 Montpellier, France, 689-696.
511

512 Jones, W. A., Ciomperlik, M. A., and Wolfenbarger, D. A. 1998. Lethal and
513 sublethal effects of insecticides on two parasitoids attacking Bemisia argentifolii
514 (Homoptera, Aleyrodidae). Biol. Control 11, 70-76.
515

516 Jones, W. A., Greenberg, S.M., and Legaspi, B. 1999. The effect of varying
517 Bemisia argentifolii and Eretmocerus mundus ratios on parasitism. Biocontrol 44,
518 13-28.
519

520 Jones, W. A., Wolfenbarger, D. A., and Kirk, A. A. 1995. Response of adult
521 parasitoids of Bemisia tabaci (Hom.: Aleyrodidae) to leaf residues of selected
522 cotton insecticides. Entomophaga 40, 153-162.
523

524 Kirk, A. A., Lacey, L. A., Brown, J. K., Ciomperlink, M. A., Goolsby, J., Vacek, D. C.,
525 Wendel, L. E., and Napompeh, B. 2000. Variation in the Bemisia tabaci s.l. species
526 complex (Hemiptera:Aleyrodidae) and its natural enemies leading to successful
527 biological control of Bemisia biotype B in the USA. Bull. Entomol. Res. 90, 317-
528 327.
529

530 Koppert Biological Systems. 2003. <http://www.koppert.nl>

531

532 Rodriguez-Rodriguez, M. D., Moreno, R., Tellez, M. M., Rodriguez-Rodriguez, M.
533 P., and Fernandez-Fernandez, R. 1994. Eretmocerus mundus (Mercet), Encarsia
534 lutea (Masi) y Encarsia transvena (Timberlake) (Hym., Aphelinidae) parasitoides de
535 Bemisia tabaci (Homoptera, Aleyrodidae) en los cultivos horticolas protegidos
536 almerienses. Bol. San. Vegetal-Plagas 20, 695-702.

537

538 Sharaf, N., and Batta, Y. 1985. Effect of some factors on the relationship between
539 the whitefly Bemisia tabaci Genn. (Homop., Aleyrodidea) and the parasitoid
540 Eretmocerus mundus Mercet (Hymenopt., Aphelinidae). Z. ang. Ent. 99, 267-276.

541

542 Simmons, A. M., and Jackson, D. M. 2000. Evaluation of foliar-applied insecticides
543 on abundance of parasitoids of Bemisia argentifolii (Homoptera: Aleyrodidae) in
544 vegetables. J. Entomol. Sci. 35, 1-8.

545

546 Simmons, G. S., and Minkenberg, O. P. 1994. Field-cage evaluation of
547 augmentative biological control of Bemisia argentifolii (Homoptera: Aleyrodidae) in
548 southern California cotton with the parasitoid Eretmocerus nr. californicus
549 (Hymenoptera: Aphelinidae). Environ. Entomol. 23, 1552-1557.

550

551 Statistical Graphics Corporation. 1999. Statgraphics Plus for Windows version 4.0.

552

553 Van Driesche, R., Hoddle, M. S., Lyon, S., and Sanderson, J. 2001. Compatibility of
554 insect growth regulators with Eretmocerus eremicus (Hymenoptera: Aphelinidae)
555 for whitefly (Homoptera:Aleyrodidae) control on poinsettias: II Trials in
556 commercial poinsettia crops. Biol. Control 20, 132-146.

557 **Figure captions**

558

559 Figure 1. Percentage mortality of Eretmocerus mundus pupae parasitizing
560 Bemisia tabaci treated with methomyl (□), indoxacarb (+), and control (□).
561 Treatments with different letters in the same day are significantly different with
562 95% of confidence, using the LSD test. Vertical bars indicate the standard error
563 of the mean.

564

565 Figure 2. Percentage mortality of Eretmocerus mundus adults treated with
566 methomyl, indoxacarb, and the control, with fresh and 24-hour old residues.
567 Treatments with different letters are significantly different with 95% of
568 confidence, using the LSD test. Vertical bars indicate the standard error of the
569 mean.

570

571 Figure 3. Percentage mortality of Eretmocerus mundus adults treated with
572 methomyl over a period of 60 days, **(A)** compared with the control treated with
573 water; methomyl (■), water (▼); and **(B)** mortality corrected using Abbot's
574 formula and with an adjusted curve ($R^2=0.88$, $P<0.01$). Vertical bars indicate
575 the standard error of the mean.

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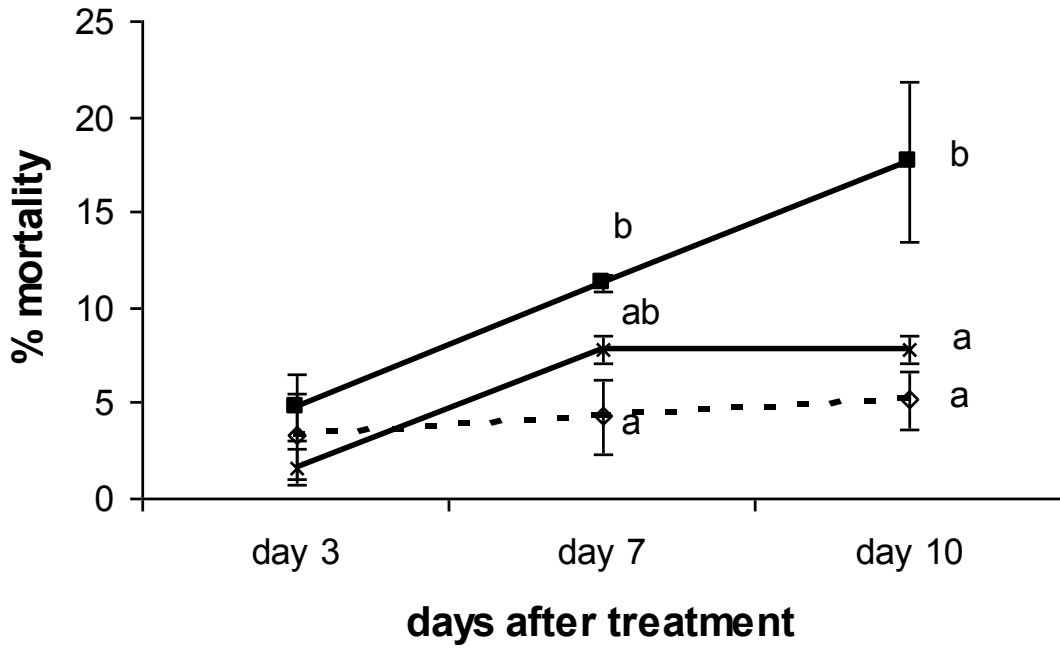
577 Figure 4. Cumulative percentage mortality of Bemisia tabaci larvae in the
578 experiment with oxamyl, **(A)** considering the four treatments, oxamyl (□),
579 oxamyl plus Eretmocerus mundus (▼), E. mundus (☆), and control (+); and **(B)**
580 considering the treatments with E. mundus (■), and without parasitoids (+).
581 Vertical bars indicate the standard error of the mean.

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583 Figure 5. Effect of oxamyl on Eretmocerus mundus parasitizing Bemisia tabaci
584 larvae. **(A)** Percentage evolution of parasitized whitefly larvae treated with
585 oxamyl (■) and water (☆); and **(B)** Cumulative percentage emergence of
586 Eretmocerus mundus adults from Bemisia tabaci larvae treated with oxamyl (■)
587 and water (☆). Vertical bars indicate the standard error of the mean.

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Figure 1

Final V

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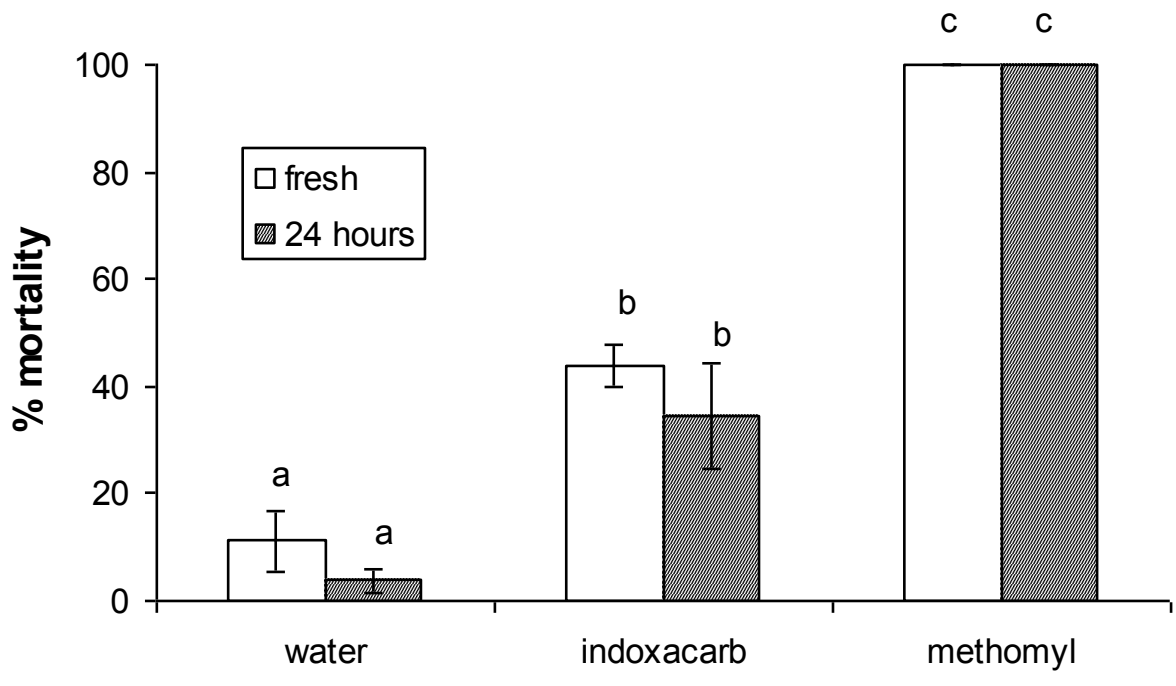


Figure 2

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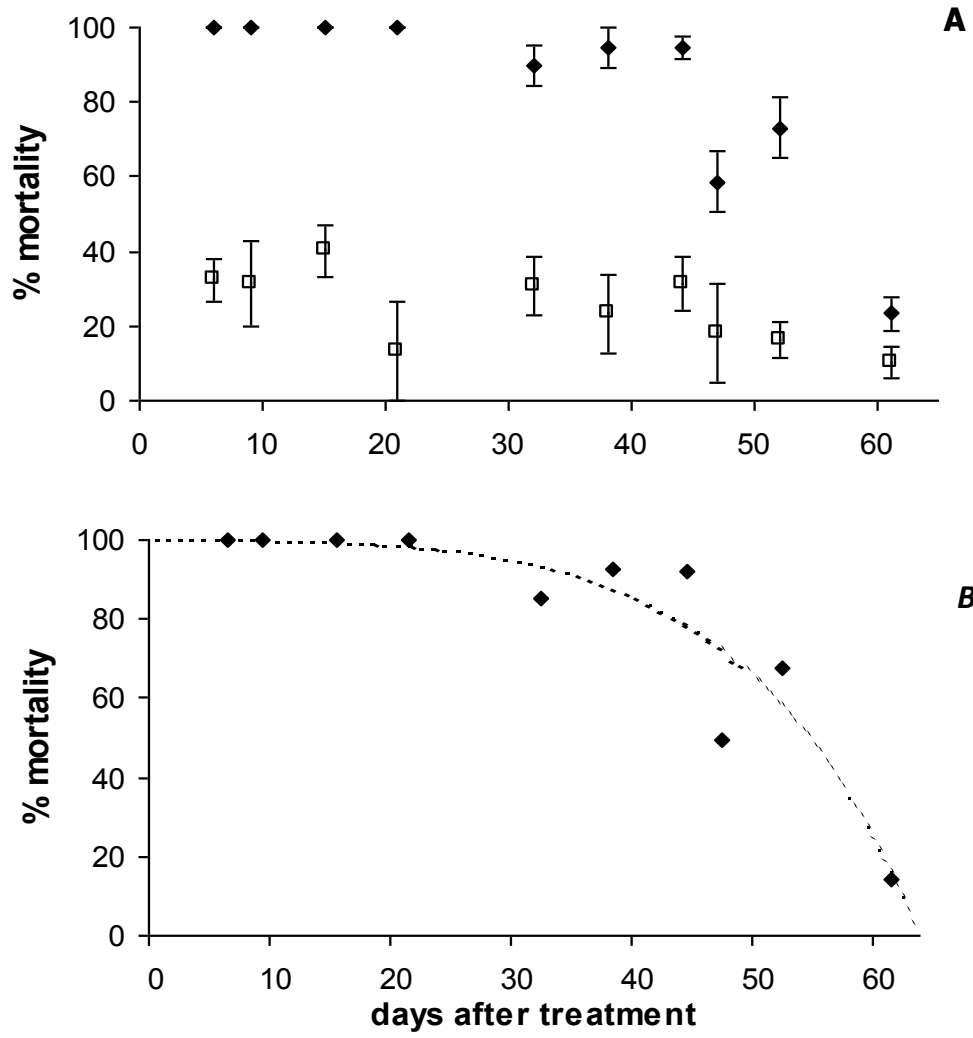
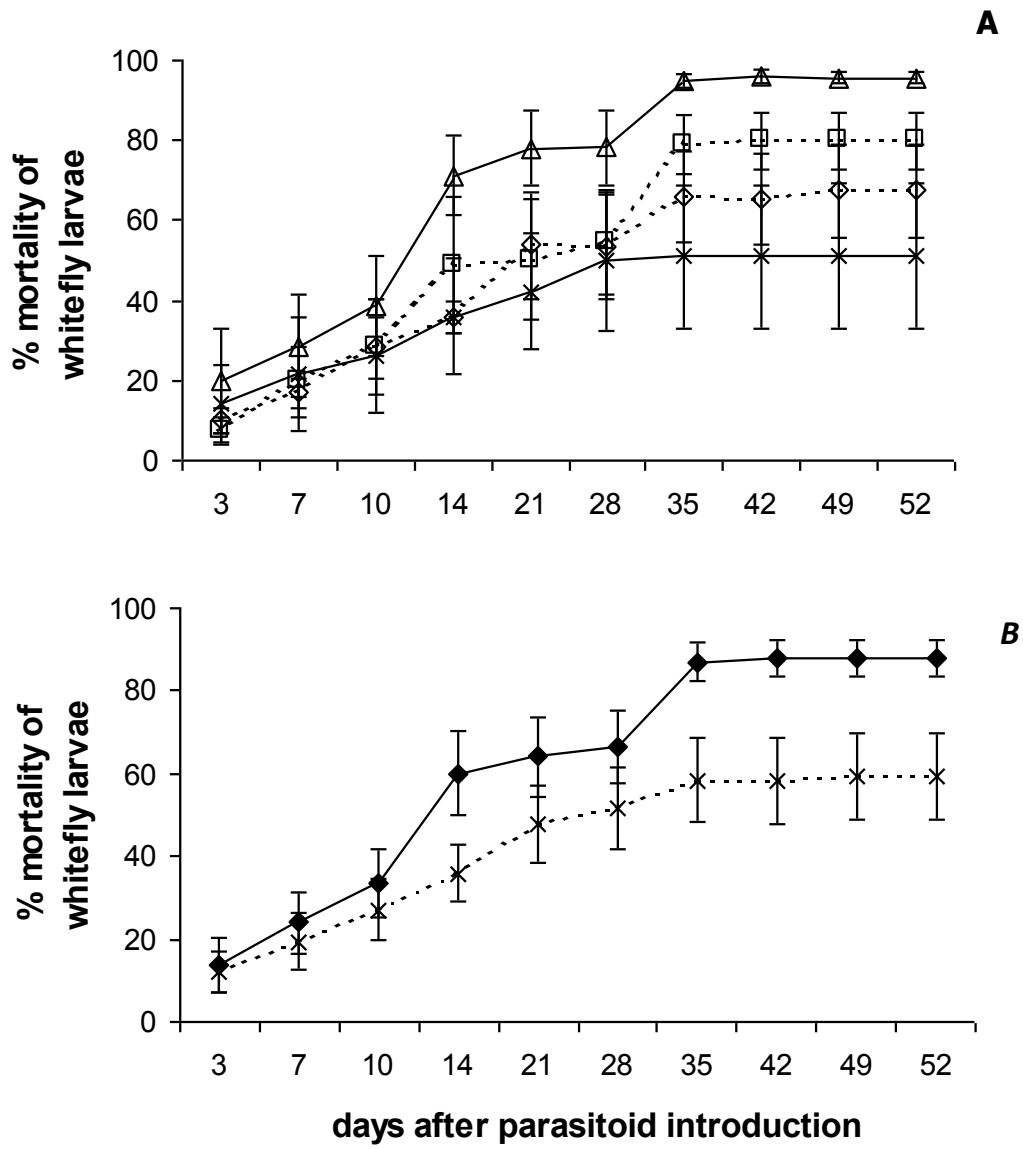


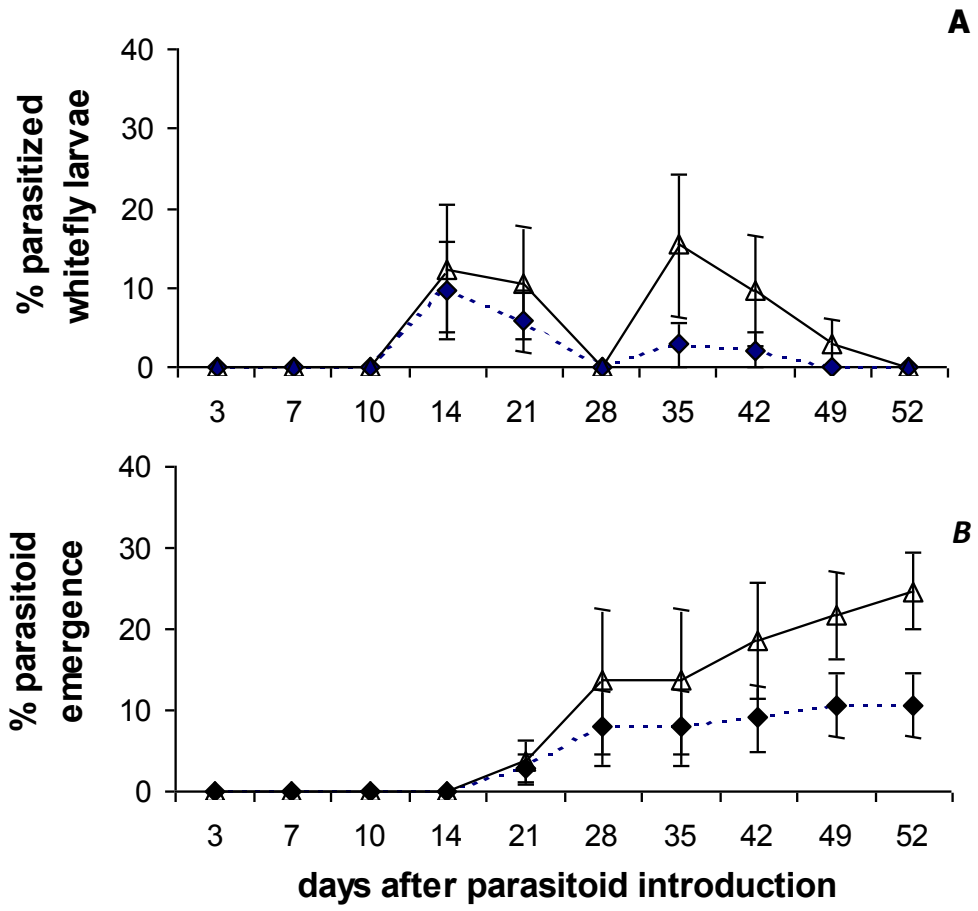
Figure 3



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Figure 4

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Figure 5