ANALYSIS OF TWO LARVAL-PUPAL PARASITOIDS (HYMENOPTERA, BRACONIDAE) IN THE BIOLOGICAL CONTROL OF *CERATITIS CAPITATA* (WIEDEMANN) IN SPANISH MEDITERRANEAN AREAS.

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Abstract: The Mediterranean fruit fly, *Ceratitis capitata* (Diptera: Tephritidae), is considered a key pest in fresh fruit and citrus production in the Mediterranean Bassin. Nowadays, it is being studied the use of several ecological methods against this pest in some Mediterranean countries (Spain, Morocco and Tunisia), like the Sterile Insect Technique (SIT), the classical biological control (CBC) and the search for native parasitoids. Two exotic larval-pupal parasitoids of fruit-flies have been imported by the IVIA to Spain: *Diachasmimorpha tryoni* and *D. longicaudata.* As it occurs in other countries, in Spain it is being studied the combined use of the SIT and overflooding releases of parasitoids to combat Medfly populations. To achieve this, only one parasitoid species is usually mass reared and released. Over the last two years we have been analyzing the competitiveness of *D. longicaudata* and *D. tryoni* in order to choose the species to be used together with the SIT for the integrated control of the Medfly.

Keywords: *Ceratitis capitata*, Biological control, parasitoid efectiveness.

Introduction

The Mediterranean fruit fly, Medfly, *Ceratitis capitata* (Wiedemann) is one of the most widespread and damaging fruit pests worldwide. Current Medfly control management in Spain is mainly based on the use of insecticides. Nonetheless, other safe environmental control strategies such as mass-trapping (Leza *et al.* 2008), chemosterilzation (Navarro-LLopis *et al.* 2007), and sterile male release (Sterile Insect Technique, SIT) (Argilés & Tejedo, 2007), among others, are being implemented since 2006 In addition, other environmentally safe control systems like Biological Control are under investigation. In the frame of two AECID projects, research groups from Spain, Tunisia and Morocco are studying the possibilities for applying the classical biological control against the Medfly by using parasitoid species of the order Hymenoptera. In this frame a survey of native parasitoids in each country was performed during 5 years, in which two pteromalid species (*Spalangia cameroni* and *Pachycrepoideus vindemmiae*) were found in Spain parasitizing naturally the medfly, only one braconid species (*Psytalia concolor*) in Morocco, and none in Tunisia (Sabater-Muñoz *et al.* 2009; Skouri, 2010; S. El Messoussi, personal communication). In the meanwhile of the survey, two braconid species (*Diachasmimorpha tryoni* and *D. longicaudata*) were imported (with relaease permits) to Spain (Sabater-Muñoz *et al.* 2009)

The main goal of this work is to compare the parasitic activity of these two species in order to choose only one of them to undertake a mass-rearing and release programme to use in combination with the undergoing SIT programme against the Medfly in Spain.

Material and methods

Adult longevity and female fertility at laboratory conditions.

Isolated couples of each species (*D. longicaudata* and *D. tryoni*) were hold in 12x12x8 cm plastic boxes (see Fig. 1), provided with water, household sugar and honey *ad libitum*. Every day 20 L3 medfly larvae were supplied through the upper mesh window, until female death. Daily mortality and fertility was analyzed for each species. Experiments were performed in a climatic chamber at 22±2ºC, 65±10% RH and 16:8 (L:D) photoperiod.

Figure 1. Plastic box used in experiments. Larvae are supplied on the top of the box mixed with the rearing media.

Effect of host density in female fertility at laboratory conditions.

Three couples of each species were tested for fertility at three different host densities (15, 60 and 120 host-larvae per box and day) and for three days, in the same boxes as described above. Total fertility produced in that period was analyzed.

Interspecific competition

The interspecific competition between the two species was studied in laboratory conditions as described above and in greenhouse conditions. At laboratory conditions, in the same plastic boxes, three couples of each species were put together and four host densities (15, 30, 60 and 120 L3 per box and day) were tested during three days. Progeny were analyzed for sex and species.

In the greenhouse, the interspecific competition was carried out in four mesh cages (120x80x60 cm) each with a citrus plant. In each cage 20 couples of both species were put together with 36, 90, 180 or 360 host-larvae per cage, during five days. Larvae were supplied to parasitoids inside five apples, var "Royal Gala", with several holes on them according to the larvae density, where we put three L3-larvae per hole, that were hold on citrus plants inside the cages (Fig. 2).

Adult longevity at year-round.

The adult longevity of both species was assessed along a year-round to cover all the climatic conditions in the Mediterranean areas of study. Each species were hold in separate, in plastic bottles of 0.9 liters with 4-5 holes for ventilation. In each bottle, 5 couples were put inside with water and food provided ad libitum. The plastic bottles were put inside wood-side mesh cages and covered with a small roof to avoid direct rain in a lemon orchard (Fig. 3). Mortality was assessed daily. Environmental conditions (temperature and relative humidity) were controlled by a data-logger.

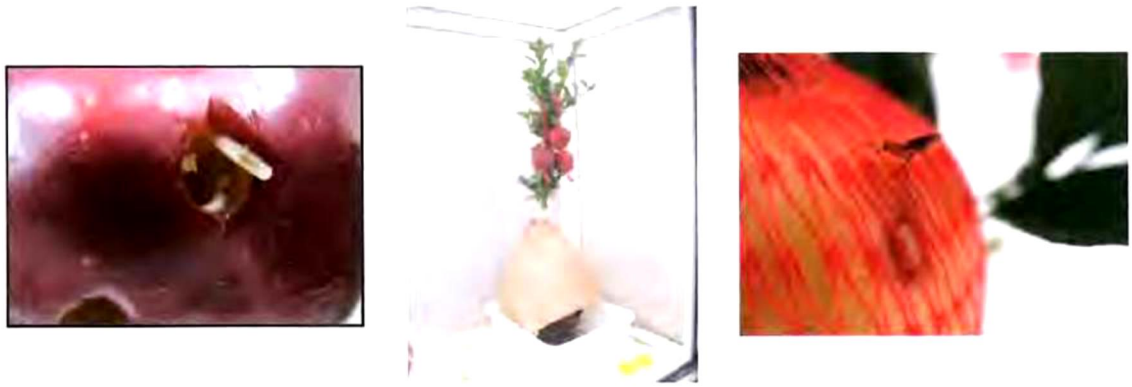


Figure 2. Introduction of larvae into apples (left), apples hanged on the plant (center) and parasitism of *D.tryoni* female in apple (right).

Figure 3. Bottles with adults inside the wodden cage at the lemon orchard.

Results and Discussion

Adult longevity and female fertility at laboratory conditions.

From the statistical analysis, the longevity of both species is different with an average of 37'2 days for *D. longicaudata* and 31'7 days for *D. tryoni*, when data about the two sexes are considered together. Whereas considering by sexes, the females of *D. tryoni* presented a significant reduced longevity (Graph 1).

Both species showed the same fertility in trials with isolated couples, with an average of 70'5 individuals per female in *D. longicaudata* and 58'2 individuals per female in *D. tryoni* (Graph 2).

Effect of host density in female fertility at laboratory conditions.

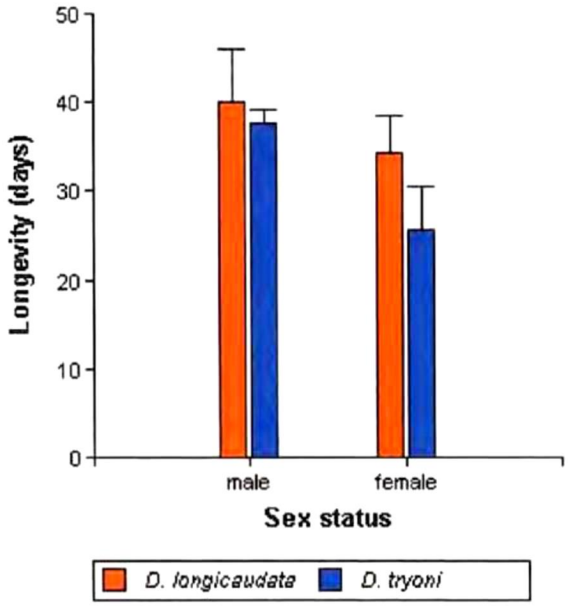
When we analyzed the fertility of females with different density of host larvae we found that it increases with the number of larvae offered (statistical differences), but there was no variation between the two species (Graph 3).

Interspecific competition

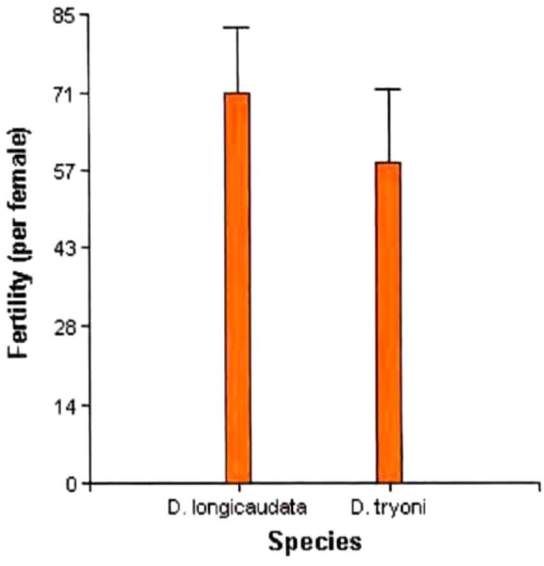
In the laboratory, the study of interspecific competition showed that with a high number of host larvae (60 and 120 per box per day) there are no statistical differences in the fertility of both species; but when the number of larvae offered is insufficient, competition is found with a predominance of *D. longicaudata* over *D. tryoni*. For *D. longicaudata* it was confirmed an increase in fertility with as increasing the density of host larvae (Graph 4).

In the field, the results shown in graph 5 confirmed the predominance of *D. longicaudata* over *D. tryoni*, as observed in laboratory. As the numbers of offered host larvae are increasing, *D. longicaudata* responded with an increase in fertility while *D. tryoni*

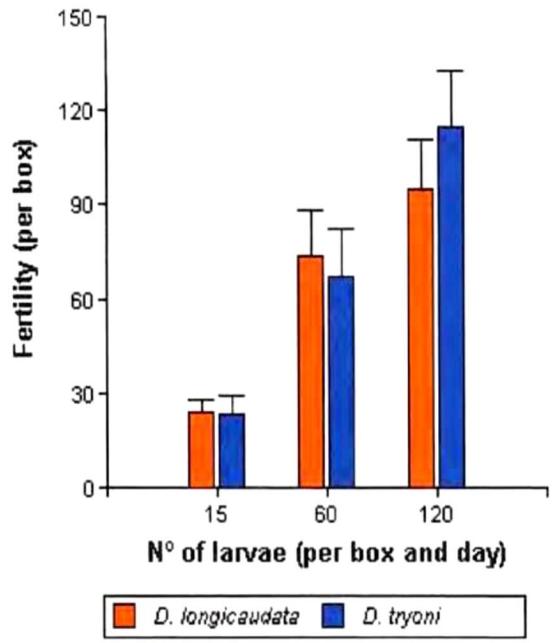
maintained in all cases a very low fertility.



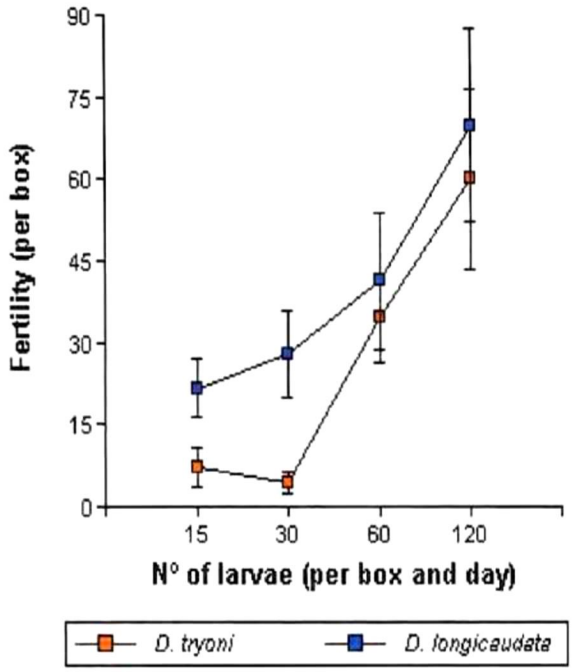
Graph 1. Adult longevity, in days, for the two parasitoid species.



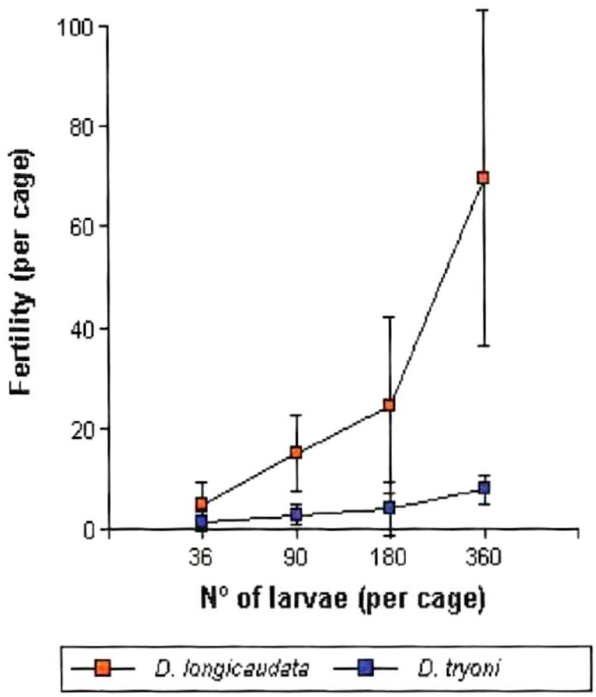
Graph 2. Average of fertility, as individuals produced by each female, in both species.



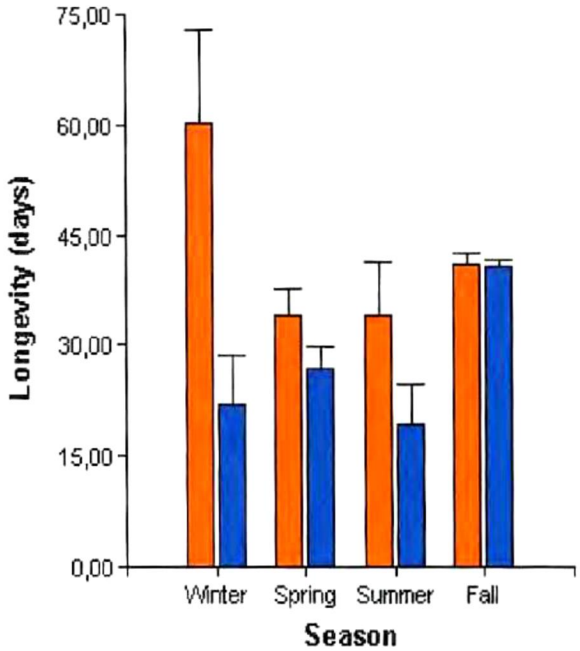
Graph 3. Average of fertility, as individuals produced per box, for the two species according to the supplied density of host larvae.



Graph 4. Average of fertility, as individuals produced per box, for the two species, in competition, according to the density of host larvae.



Graph 5. Average of fertility, as individuals produced per cage in the two species according to the density of host larvae.



Graph 6. Longevity, in days, for adults of the two parasitoid species in the four year-seasons at field conditions.

Adult longevity at year-round.

In Graph 6 we can see the results on adult longevity through the year. There were statistical differences in longevity between the two species for all the year seasons, except in Fall period, where total adult mortality was reached in the two species at the same time due to below 0°C temperatures. In the other three seasons, *D. longicaudata* present a significant higher longevity.

Conclusions

D. longicaudata seems to present a higher performance than *D. tryoni* for the parasitism on *C. capitata* in Mediterranean conditions. The results support the view that *D. longicaudata* is the most suitable species to be included in the implementation of biological control in the Comunidad Valenciana, complementing the use of the SIT against the pest. This species, *D. longicaudata*, could also be considered for its use in Northern Morocco and Tunisia due to the similar climatic conditions in these countries to those presented in this work.

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