# Rearing *Fopius arisanus* (Sonan) (Hymenoptera: Braconidae) in Mediterranean Fruit Fly (Diptera: Tephritidae)

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**Abstract.** In Hawaii, the egg-larval parasitoid *Fopius (=Biosteres) arisanus* (Sonan) (= *Opius oophilus* Fullaway) (Hymenotera: Braconidae) is the most effective parasitoid of its preferred host the oriental fruit fly, *Bactrocera dorsalis* Hendel (Diptera: Tephritidae). Other Hymenopterous parasitoid species introduced into Hawaii still exist but vary considerably in their distribution and effectiveness as biological control agents. Based on field observations of *F. arisanus* parasitizing medfly in coffee and oriental fruit fly in guava, we postulated that a strain of *F. arisanus* could be selected in the laboratory to be reared exclusively on medfly. We report the results of these studies.

**Key words:** parasitoid, oriental fruit fly, medfly, parasitoid host adaptation

## Introduction

The Mediterranean fruit fly, (medfly) *Ceratitis capitata* (Wiedemann) Diptera: Tephritidae) has worldwide distribution and is one of the most destructive tephritid pests of deciduous and subtropical fruits (Back and Pemberton 1918). Growing concerns over unwanted risks associated with the use of pesticide chemicals have provided a mandate to explore the potential of other fruit fly control strategies that are safe and ecologically non-disruptive. Biological control is a safe and viable alternative that satisfies such requirements. At the USDA-ARS Laboratory in Honolulu, HI, we rear *Fopius arisanus (Biosteres arisanus* Sonan) (= O*pius oophilus* Fullaway) on the oriental fruit fly (Harris et al. 1991), *Bactrocera* (*Dacus dorsalis*) (Hendel), the parasitoid's natural host. One of the two known opiine egg parasitoids, *F. arisanus* is abundant in the Hawaiian agroecosystem (Bess 1953; Bess et al. 1961). Thus, introduction of *F. arisanus* into habitats where medfly is the target host could be beneficial to the success of a classical parasitoid release program and offer a new option for Integrated Pest Management of medfly by augmentatative parasitoid releases.

In entomophagous insects, the selection of host-adapted strains is still unexplored, yet considered a promising research area (Zenil et al. 2004). Notwithstanding, we are attempting to raise a colony of medfly adapted strain of *F. arisanus*. If successful, this may offer a wide range of new possibilities in places where effective parasitoids of medfly are conspicuously absent. Zenil et al. (2004), demonstrated adaptation of *F. arisanus* to medfly and to Anestrepha spp in Mexico documenting the feasibility of this approach. The work of Quimio and Walter 2001 showed *F. arisanus* successfully parasitized *B. tryoni*, *B. jarvisi* and *B. cucumis* in Australia further corroborating the capability of *F. arisanus* to parasitize tephritid fruit flies.

## Materials and Methods

Our approach consisted of rearing *F. arisanus* sequentially on medfly with expectation of increasing the level of parasitoid adaptation from physiological conditioning with each generation. Based on the work of Harris et al. (1991) and Bautista et al. (1999), we hypothesized that medfly adaptation could be greatly accelerated by laboratory selection.

*Fopius arisanus* was obtained from a colony of parasitoids maintained on oriental fruit fly for ca. 200 generations. For this experiment, oriental fruit fly and medfly eggs were produced from flies reared as larvae on wheat diet (Tanaka el. 1969). Assays were conducted in the laboratory with temperature of 25°C, 69% relative humidity and continuous lighting (24 h).

**Biological attributes of medfly-reared parasitoids.** Preimago (number of days from egg oviposition to adult emergence) in medfly reared parasitoids (FACC) was compared to those of oriental fruit fly reared parasitoids (FABD) (Fig 1). The duration of preimago development was determined from daily emergence of parasitoids in puparial cohorts. The other biological parameters (preimago, progeny/female, adult longevity, and percent female progeny) were quantified from 100 pairs of parasitoids fed with honey and water. Ripe papayas, *Carica papaya* L. were trimmed into sections (8 X 4 x 1.3 cm) and perforated with about 20 holes (5 mm deep), with a camel hair brush. Each fruit was inoculated with 2,000 freshly laid medfly or oriental fruit fly eggs. Inoculated fruit was exposed to parasitoids inside a cubical cage (30 X 30 X 30 cm) for 24 h. The females in the cubical cages were permitted to lay eggs every 2 days until 95% of reproducing females had died. Mortality of parasitoids was recorded daily.

Percent parasitization was determined by individually dissecting 100 eggs per 2 day laying period. The remainder of eggs in fruit sections hatched and were reared on wheat diet (Tanaka et al. 1969) until emergence of parasitoids (Bautista et al. 1999). Preimago development tests were replicated 5 times using fresh batches of hosts and parasitoids each time.

**Trend in generational productivity of experimental colony.** Improvement of parasitoid laboratory production and adaptation was obtained rearing the parasitoid sequentually generation by generation.

Ripe papayas, *Carica papaya* L., were trimmed into sections as described in the previous section. Approximately 5,000 freshly laid medfly eggs (collected within 24h) were inoculated into the fruit by inserting 250 eggs per hole. As many as 5 inoculated fruits were exposed to ~4–5,000 parasitoids inside a rectangular cage (61 X 41 X 32 cm). Egg laying by parasitoids commenced when females were 8–10 days old posteclosion. Thereafter, eggs were collected from females every other day for 2 weeks from 5–8 breeding cages simultaneously. Fruits were recovered after exposure to parasitoids for 24 h. Fruits were placed on medfly rearing diet (Tanaka et al. 1969) where the hosts completed larval development.

Puparia recovered from parasitoid females which laid their eggs on the same dates were pooled and weighed. A 2-g sample was taken from each puparial batch to generate data on puparial count and parasitoid emergence. Mean percent parasitoid yield per generation was expressed as ratio between total number of emerged parasitoids and total puparia recovered. The remainder of puparia was returned to breeding cages to produce parasitoids for the next generation.

**Reciprocal cross parasitization of medfly and oriental fruit fly-reared parasitoids.** Using the rearing methods previously described, biological parameters generated were quantified by comparing progeny production and sex ratio of FACC parasitoids reared on medfly and switched to oriental fruit and FABD parasitoids reared on *B. dorsalis* and switched to medfly. This information would be beneficial for understanding reciprocal cross parasitization of medfly and B. dorsalis, which occurs in the field in Hawaii.

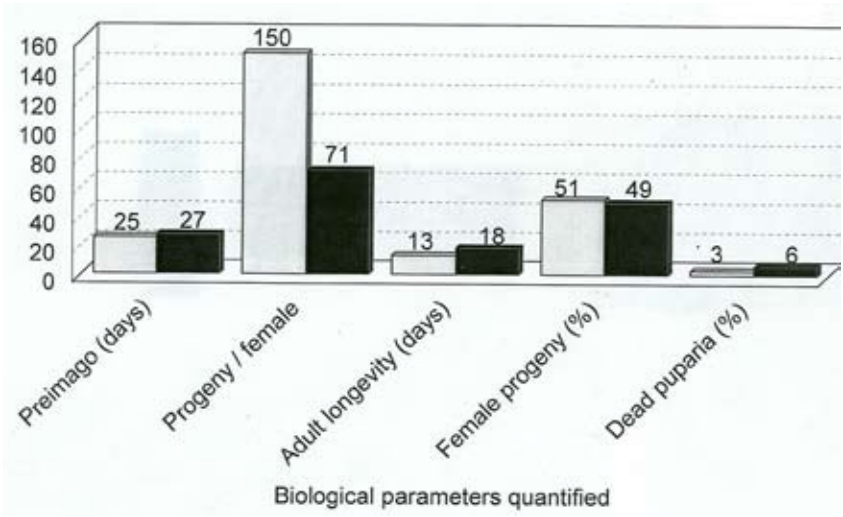


**Figure 1.** Attributes of *F. arisanus* preimago development compared between FACC and FABD.

**Statistical analysis.** Data from each experiment were analyzed by analysis of variance (ANOVA) using SAS GLM procedure from the SAS System for Windows Version 7.0 1998 by SAS Institute Inc., Cary, NC, USA. Means were separated by GLM LSD.

## Results

Biological Attributes of Medfly-Reared Parasitoids: Dissection of parasitized eggs (F12–F14) showed that both FACC and FABD females commenced to lay eggs in fruit fly eggs within 24 hours after emergence. However, FACC females continued to oviposit in medfly eggs up to 36 days post eclosion or 12 days longer than FABD (Fig 3). FACC female mean oviposition period was 8 days longer than FABD females. Gravid females in FACC (as early as F15) produced half as many progeny as FABD females. This corresponds to a mean total progeny of 72 (range = 68–87) produced by a FACC female in her lifetime compared to 150 progeny by FABD on oriental fruit fly. Female progeny sex ratio of 49% in FACC (range = 42–51%) was comparable to a female sex ratio of 51% in FABD (range = 45-56%).

FACC adults lived longer than FABD adults with males and females surviving 5 and 9 d longer than those of FABD, respectively. The proportion of dead oriental fruit fly or medfly puparia that resulted from parasitization by *F. arisanus* was as low as 10%. Although the longevity of FACC adults was higher than FABD adults the difference was not significant ($n = 26$, $F = 1.36$, $p = 0.248$, $df = 1, 59$).

**Trend in generational productivity of experimental colony.** Fig. 2 shows the trend in parasitoid productivity as rearing on medfly (FACC) progressed from parental (P) to (F25) generation. For comparison purposes corresponding yield data from cohorts of FABD are presented. Substitution of *B. dorsalis* with medfly eggs as host of *F. arisanus* resulted in mean progeny yield of only 10% in the P generation compared to 67% of FABD. The drastic effect of the host switch on the yield performance of the test culture was evident in the first few generations (F1–F8) with recovery rates oscillating between 2 and 9%. This evidence

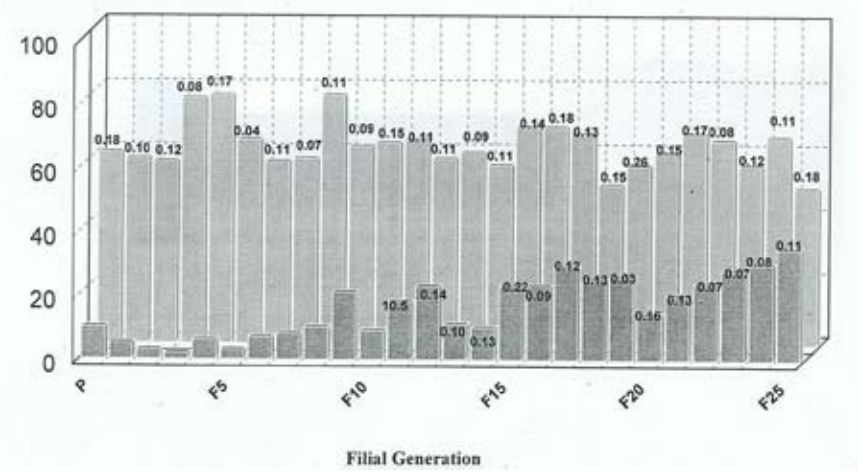


**Figure 2.** Generational trend in the productivity of FACC. Standard deviation shown at top of columns.

suggests that only a few *F. arisanus* females oviposited in FACC eggs or egg hatch and survival was low. In F9, a few more females contributed to the production of progeny. The yield increased and gradual improvement continued thereafter. Interestingly, parasitoid yield fluctuated between peaks in F9, F12, F16 and F21. The reason for this variation is the parasitoid culture was going through the bottle neck of adapting to the new (FACC) host. During this study, a low number of FACC females produced parasitoids in the beginning but the yield increased to a mean progeny yield of ca. 40% in the F25 generation (Fig 2). The same pattern of increase in parasitoid progeny yield by generation was experienced in the development of the FABD colony. Measurement variables were calculated from 760 individuals of FACC and 524 of FABD and compared with SAS GLM LSD. The results, as expected, showed that FABD parasitoid yield was significantly higher than FACC parasitoid yield ($n = 26$, $F = 359.77$, $p = 0.0001$; $df = 1, 50$). The pattern of increase in FACC yield generation by generation showed that colony establishment was assured.

**Reciprocal cross parasitization of medfly and oriental fruit fly.** Data was summarized on the results of reciprocal cross rearing of *F. arisanus* on medfly and then on oriental fruit and rearing of *F. arisanus* on oriental fruit fly and then on medfly. The results showed that when oriental fruit fly is used as the origin host and rearing host of *F. arisanus*, progeny production of the parasitoid was lowest (16). When medfly was the origin host, oriental fruit fly progeny production was highest (203). When oriental fruit fly was used as the origin host and rearing host progeny production was high (189). When medfly was used as the origin and rearing host, *F. arisanus* progeny production was low (65). When *B. dorsalis* was used as the rearing host, *F. arisanus* parasitoioid progeny production is high. These preliminary results suggest that after host switching is accomplished rearing *F. arisanus* on medfly, it is better to continue production of the parasitoid on medfly. Switching back to *B. dorsalis* could result in lower parasitoid progeny production. Therefore *F. arisanus* adapted to medfly or to *B. dorsalis* should be maintained in separate colonies in the laboratory. Further study is necessary to verify these preliminary results.

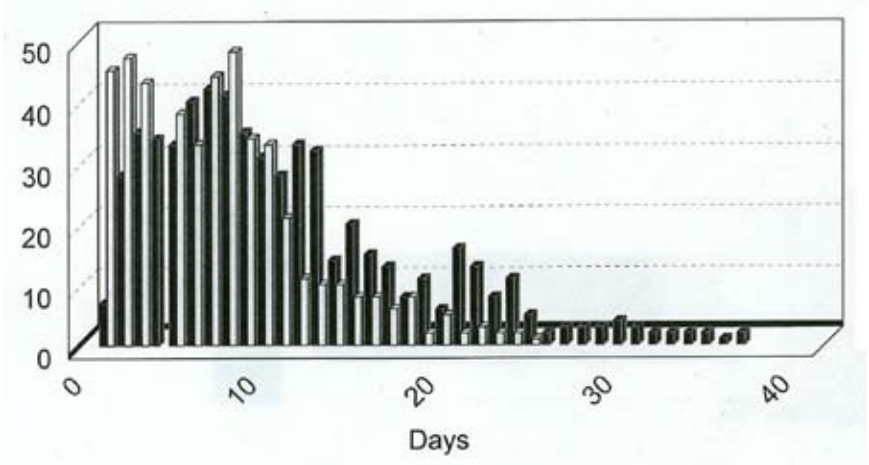


**Figure 3.** Percent parasitism / 100 eggs dissected showing different ppropensity for oviposi tion by female FAB (open bars) and FACC (black bars) .

## Conclusions

We demonstrated experimentally the feasibility of establishing a colony of *F. arisanus* on medfly by sequential rearing resulting in adaptation to medfly. This accomplishment increases possibilities for successful introduction and establishment of *F. arisanus* into habitats where medfly is the target pest. Successful development of the medfly-adapted strain *of F. arisanus* should facilitate transfer of this parasitoid and its rearing technology to countries where medfly is the only available host. As a biocontrol agent, a medfly-adapted strain *of F. arisanus* could compliment the Sterile Insect Technique and other compatible control strategies in the Integrated Pest Management of *C. capitata*. The medfly adapted strain of *F. arisanus* provides a new tool for augmentative parasitoid releases to control medfly and oriental fruit fly in area-wide IPM programs in Hawaii and elsewhere

## Acknowlegement

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