Field Release of *Fopius ceratitivorus* (Wharton) (Hymenoptera: Braconidae), for Biological Control of medfly, *Ceratitis capitata*, in Hawai`i

Draft Environmental Assessment

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I. Proposed Action

A. Summary

The University has submitted an application to the Plant Pest Control Branch, Hawaii Department of Agriculture (HDOA), to the HDOA Plant Quarantine Branch, 1849 Auiki Street, Honolulu, HI 96819 for a permit to release ***Fopius ceratitis* (Hymenoptera: Braconidae)** into the environment of the State of Hawai`i under the provisions of Hawaii Revised Statutes, Chapter 141, Department of Agriculture, and Chapter 150A, Plant and Non-Domestic Animal Quarantine an application to the Hawaii Dept. of Agriculture, Plant Quarantine Branch (DOA-PQB). Applicant is asking for a permit and seeking to release from quarantine the host specific parasitoid *Fopius ceratitivorus* Wharton (Hymenoptera: Braconidae) for enhanced biological control of the Mediterranean fruit fly, *Ceratitis capitata* (Diptera: Tephritidae) in coffee and other cropping systems throughout Hawai`i.

This Draft Environmental Assessment (DEA) was prepared by the applicant for the Office of Environmental Quality Control (OEQC), Department of Health, State of Hawaii, to comply with the provisions of Hawai`i Revised Statutes, Chapter 343, Environmental Impact Statements.

B. Identification of Applicant

The University of Hawai`i is the project applicant.

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C. Regulatory Authority

The University of Hawai`i is the approving agency.

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D. Anticipated Determination

Finding of no significant impact (FONSI)

II. Need for the Proposed Action

A. Detailed description of proposed action

We propose to release the African parasitoid *Fopius ceratitivorus* in Hawaii's coffee plantations and other crops for control of the Mediterranean fruit fly.

B. Need for the release

The target pest, the Mediteranean fruit fly (*Ceratitis capitata*) is one of the most important agricultural pests in the world, infesting hundreds of species of fruits and vegetables (Liquido et al. 1991). In Hawaii, it is important both as a direct pest and as a quarantine pest of crops such as citrus, eggplant, guava, loquat, mango, melon, papaya, passion fruit, peach, pepper, persimmon, plum, star fruit, tomato, and zucchini. Current control practices for medfly (promulgated by the USDA-ARS-PBARC Area-Wide Fruit Fly Integrated Pest Management Program) rely on a combination of pesticide-treated bait sprays and field sanitation; as well as the sterile insect technique (SIT); the release of mass-reared extant parasitoids; and semio-chemical based male annihilation (Vargas 2005). The sustainability of the latter three techniques, once the Federal government stops the influx of Area-Wide implementation funding, is questionable. The use of GF-120 as a bait spray is safer than the previous alternative (malathion); but it has nevertheless been shown to be toxic to a wide array of beneficial (Wang et al. 2005) and other non-target insects (Wang & Messing 2006).

Numerous entomologists have emphasized the importance and potential economic benefit of introducing new parasitoids of tephritid fruit flies into Hawaii and other infested regions (Gilstrap & Hart 1987, Greathead & Waage 1983, Messing 1995, Steck et al. 1986, Wharton 1989). Biological control is increasingly viewed as a practical, safe, and economically effective means of fruit fly control, and its importance continues to grow as pesticide use becomes more restricted. Imported parasitoids can incrementally increase fly mortality, reduce infestations, and contribute to a systems approach to quarantine security for fruit and vegetable industry exports (Jang & Moffit 1994).

Locations of rearing facilities and release sites

Current rearing:
Hawaii Dept. of Agriculture Quarantine Facility
1428 S King St., Honolulu, HI 96814-2512

University of Hawaii at Manoa
Gilmore Hall- 3050 Maile Way
Honolulu, HI 96822

Initial releases to be made at:
Kauai Agricultural Research Center
7370 Kuamoo Rd, Kapaa, HI 96746

Number/Quality to be released
We estimate in the range of several thousand adult parasitoids release in coffee orchards.

Timing of release

Spring to summer of 2009.

Method of release

Adult parasitoids will be collected in vials in the rearing room, and the vials will be placed under medfly-infested coffee plants for the wasps to emerge.

Common Name and Scientific Classification
Fopius ceratitivorus (Hymenoptera: Braconidae)

Location of Voucher specimens

Voucher specimens are deposited in the Hawaii Department of Agriculture Taxonomy Unit (1428 South King Street, Honolulu, Hawaii 96814-2512); and also at the Texas A & M University Insect Collection, College Station, Texas.

B. Information on the target (host) organism

Classification of target (host) organism

*Ceratitis capitata (*Weidemann): Diptera: Tephritidae

common name = medfly, or Mediterranean fruit fly.

Life history of the target organism

The entire life cycle takes 30–50 days. A female lays 1–10 eggs in a fruit, and may lay as many as 22 eggs per day, and as many as 800 eggs during her lifetime (normally 300–500). Usually females die soon after they cease to oviposit. Under optimum conditions, Medfly can complete its life cycle, which consists of four stages (adult, egg, larvae, and pupae) within 30 days. At lower temperatures, Medfly requires longer time intervals of up to 100 days to complete its life cycle. The eggs, laid just under the skin of the susceptible fruits, hatch within a few days and the emerging maggots or larvae feed on the fruit pulp. This is the point at which economic damage occurs. The maggots are fully grown, about one centimeter long, within 7-24 days. When mature, they make their way to the surface of the fruit, drop to the ground, tunnel into the soil and pupate. The adult fly is formed within the pupa and emerges within 8-46 days, forcing its way to the surface of the soil. The newly emerged adults require about 2-3 days to mature before starting to lay eggs.

Pest status of the target organism

The medfly is a major pest of fruits and vegetables in Hawaii, where it attacks avocado, banana, bittermelon, carambola (star fruit), coffee, guava, mango, orange, papaya, peppers, persimmon, and more. Besides being a direct pest causing crop losses, the medfly contributes to increased farm production costs, increased use of toxic insecticides, and quarantine restrictions on horticultural exports. Fruit flies are consistently mentioned in commodity group industry analyses as a major impediment to agricultural growth and diversification

C. Biology of the organism to be released

Fopius certatitivorus (Hymenoptera: Braconide) Life History

Fopius ceratitivorus has a typical opine braconid koinobiont life history. The adult female wasp oviposits into eggs of medfly within fruit tissues, and the wasp larvae develop inside the host, eventually killing the flies in the puparium.

Parasitoid developmental time from egg to adult is 21.8 days (Bokonan-Ganta et al. 2005). Mean longevity of ovipositing females is 16.2 ± 0.5 d; ovarian maturation peaks at 61.6 mature eggs per female on the fifth day after eclosion. Mean number of offspring produced per day by mated females is 5.1 ± 0.4, and realized fecundity is 107.8 ± 12.8 eggs deposited during the female's lifetime (Bokonan-Ganta et al. 2007).

In Guatemalan coffee plantations, field releases of *F. ceratitivorus* resulted in 50-60% reduction of medfly pupae (Pedro Rendon, personal communication).

We cannot measure dispersal capability, since the parasitoid is not allowed out of quarantine. Economic and political considerations make long-term studies in the area of origin (central Kenya) unfeasible.

Natural Geographic Range of *Fopius certatitivorus*

Fopius ceratitivorus is known only from central Kenya, where it was collected in highland coffee plantations near the towns of Ruiru (1°5.72'S, 36°54.22'E at 1609 m elevation) and Rurima (0°38.39'S, 37°29.69'E at 1228 m elevation). Mean annual rainfall in these areas are 1.06 m and 0.9 m, respectively, and the mean temperature ranges are 13-25°C and 15-28°C, respectively (Wharton et al. 2000).

Host range of *Fopius certatitivorus*

Ceratitis capitata (Wiedemann) is the only known host of *Fopius ceratitivorus*.

Host Range List

Ceratitis capitata

Parasites/hyperparasites

None.

Status as hyperparasite

Fopius certatitivorus does not act as a hyperparasite

III. Alternatives to the Proposed Action

The actions being considered in this DEA are (1) No Action (no release) or (2) release. The No-Action alternative would be an acceptance of the status quo of infestation of the Mediterranean fruit fly in Hawaii. This leads to the continued use of large amounts of chemical insecticides in the Islands, with their threat to non-target organisms, human health, and groundwater contamination. It also means continued farm losses to medfly, including direct crop loss, increased production costs, and loss of markets.

IV. Environmental Impacts of the Proposed Action and Alternatives

Expected environmental impacts of the proposed release

There are two main issues contributing to the risk/benefit analysis for release of *F. ceratitivorus*: (1) environmental safety (= risk), and (2) potential efficacy against medfly (= benefit).

There are 33 potential non-target tephritid fly species in Hawaii, including 26 endemic species, and five deliberately introduced and two inadvertently introduced weed biocontrol agents (Hardy and Delfinado 1980). Among the 26 endemic species, 21 species belong to the genus *Trupanea*, which are predominantly flower-head feeders. The other 5 are stem miners. **Not a single one of these species feed on or in fruits.**

In quarantine, we tested the potential impact of *F. ceratitivorus* against several representative non-target flies in Hawaii, including the gall-forming weed biocontrol agent *Procecidochares alani* and (following the recommendations of the Hawaii Dept. of Agriculture Plants and Animals Advisory Committee) the endemic flowerhead feeding fly *Trupanea dubautiae*, as well as another gall-forming weed biocontrol fly, *Eutreta xanthochaeta*. These were chosen to represent both native and exotic non-target flies; and feeders in both types of plant tissue (flowers and stem galls). There are no non-target fruit feeders in Hawaii.

Results of these studies have been published in peer-reviewed scientific journals (the test with the lantana gall fly has been submitted and is currently in review). The manuscripts are attached in support of this application (Attachments 4, 5, and 6). Using recognized testing protocols, **not a single case of successful parasitism of a non-target fly was recorded.** Representative data are shown in the tables below. The egg-attacking parasitoids simply do not recognize non-target flies as suitable hosts – they do not even probe (i.e., try to sting with their ovipositor) into the substrate.

Table 1: Behavioral responses of three *Fopius* parasitoids to the endemic flowerhead-feeding fly. *Trupanae dubautiae.*

Test parasitoids	Choice test		No-Choice test	
	Frequency of probes on		Frequency of visits to flowerheads	Frequency of probes into flowerheads
	Papaya fruit	Flowerheads		
Fopius arisanus	26.2 ± 4.2	0	6.1 ± 1.8	0
F. caudatus	14.8 ± 1.0	0	4.4 ± 1.5	0
F. ceratitivorus	7.7 ± 2.3	0	2.6 ± 0.3	0

Table 2: Behavioral responses of *F. ceratitivorus* to *Lantana* plants containing lantana gall fly eggs and larvae.

Test condition	Frequency of visits to:		Frequency of ovipositor probes in:	
	Lantana galls	Papaya fruit	Lantana galls	Papaya fruit
No-choice	0.00 ± 0.00	-	0.00 ± 0.00	-
Choice	0.00 ± 0.00	9.75 ± 0.25	0.00 ± 0.00	7.25 ± 0.48

The host range of *F. ceratitivorus* is so narrow that, not only is it unable to parasitize non-target flies, it cannot even parasitize three more closely related pest fruit flies in Hawaii (*Bactrocera cucurbitae, B. dorsalis,* and *B. latifrons*) (Bokonon-Ganta et al 2005). Thus *F. ceratitivorus* is more host-specific than all previously introduced fruit fly parasitoids in Hawaii except *Psyttlia fletcheri*. None of these other parasitoids with broader host ranges that have been established here for decades have had any significant environmental impactss in Hawaii.

Table 2: Percent parasitism of four fruit fly species exposed to *F. ceratitivorus* in laboratory cage tests.

Fly species	No. of hosts	Parasitism (%)[1]		
		Egg	First instar	Second instar
B. cucurbitae	125.8 ± 9.5 a	0 (8)	-	-
B. dorsalis	128.3 ± 9.8 a	0 (7)	-	-
B. latifrons	123.7 ± 7.6 a	0 (6)	-	-
C. capitata	128.3 ± 13.1 a	10.0 ± 0.9 (12)	0.6 ± 0.5 (6)	0 (5)

[1] Numbers in parentheses are the number of replicates. Means followed by the same letters are not significantly different ($p > 0.05$).

Based on these findings, we are confident that the risk of environmental effect from introducing this parasitoid to Hawaii is extremely low.

Potential impacts on human environment

There will be no anticipated impacts of the release of *F. ceratitivorus* on the human environment in Hawaii. A positive impact would be the reduction of current control practices for medfly, which require pesticide treatments that may prove hazardous for human populations.

Literature search for other host records

Medfly is the only known host of *F. ceratitivorus.*

Host specificity in country of origin

Medfly is the only known host of *F. ceratitivorus.*

Interactions with established biocontrol agents

We plan to release *F. ceratitivorus* to augment the effectiveness of *F. arisanus,* which is already established in Hawaii. Studies of competition with extant parasitoids revealed that *F. ceratitivorus* and *F. arisanus* have the same chance of winning in intrinsic competition with one another, depending on which one occupies the host first (Bokonon-Ganta et al. 2005). *F. ceratitivorus* is expected to do better than *F. arisanus* at higher elevations and cooler sites where the latter species is absent or relatively rare (Kroder & Messing, in prep).

It's been widely demonstrated that a complex of introduced natural enemies usually provides greater levels of biological control than those provided by individual agents (Huffaker et al. 1976, Clausen 1978, Greathead 1986, Bokonon-Ganta et al. 1996). Also, based on analytical models (i.e., May & Hassell 1981) it's been shown that the introduction of a second parasitoid species generally enhances host depression or, at most, does not alter it.

Potential impacts on threatened and endangered species

There are no anticipated impacts to threatened and endangered species.

Impact to related non-target potential hosts

There are no anticipated impacts to related non-target potential hosts.

Potential of *F. ceratitivorus* to act as a hyperparasite

F. ceratitivorus does not act as a hyperparasite.

Potential of *F. ceratitivorus* to attack non-targets

The risk assessment for potential impact of *F. ceratitivorus* on non-targets can be addressed by three complementary approaches: theoretical, empirical, and experimental.

Theoretical context.

From a behavioral ecology standpoint, numerous studies on host finding and host acceptance behavior of braconids show that micro-habitat selection plays a key role in host selection (Nishida 1956, Messing et al. 1996). Visual and olfactory stimuli (e.g. fruit shape, size, color, and odors) are critical to habitat recognition by parasitoids attacking fruit-inhabiting flies (Greany et al. 1977, Vargas et al. 1991, Leyva et al., 1991, Messing & Jang 1992, Baustista & Harris 1996). Natural microhabitats recognized by parasitoids include host-infested fruit (often associated with olfactory stimuli from chemical changes associated with infestation) and, to a lesser extent, fruit-shaped galls. The parasitoids do not recognize stems and flowers as suitable host-habitats, so they do not search there, and they do not oviposit there. Habitat serves as a filter to restrict the range of possible hosts at risk.

Not a single endemic tephritid, nor any of the deliberately introduced tephritids, utilize fruits as a host.

Empirical record.

Extensive field surveys in Hawaii show the following:

(1) Seven endemic flowerhead-feeders in the genus *Trupanea* and one endemic gall-former (*Phaeograma sp.*) were surveyed on Kauai and Maui (Duan et al. 1996, Duan & Messing 1998; John Herr, unpublished). Not a single deliberately introduced fruit fly parasitoid was recovered from any of these flies.

(2) Surveys on introduced weed control flies (3 gall formers) were conducted by Bess & Haramoto (1959, 1972), Wong et al. (1991), and Duan et al. (1996). Bess & Haramoto state that

the two introduced opiines with the broadest host range, *D. longicaudata* and *D. tryoni*, are "*casual parasites*" of *P. utilis*. i.e., the fly is only attacked when the parasites are abundant and when their normal hosts (medfly and Oriental fruit fly) overlap in distribution with the gall flies. In collections of tens of thousands of galls of Maui pamakani, less than 1% were attacked by opiines. Previous records stating parasitism of *P. alani* by *D. tryoni* (Funasaki et al. 1988) were subsequently proven to be false (Purcell et al. 1977).

(3) *D. tryoni* and *D. longicaudata* parasitize the lantana gall fly, *Eutreta xanthochaeta*, on *Lantana camara*. At its maximum, *D. tryoni* was found to cause 10% indispensable, mortality of local populations of the lantana gall fly (Duan et al. 1998). The gall fly itself has never been shown to be of value in controlling lantana.

(4) A survey on Kauai showed that no deliberately introduced fruit fly parasitoids were recovered from field-collected *Ensina sonchi* larvae feeding on flowers of the introduced weed, *S. oleraceae* (Duan & Messing 1998).

(5) Additional surveys in Kenya (where *F. ceratitivorus* originated) likewise showed no parasitism of flower head feeding flies (Trostle 2005).

Together, these data suggest that the only significant impact of the many opiine braconid parasitoid species introduced to Hawaii over the years has been two larval parasitoids on the gall forming *Eutreta (*and 10% mortality is much less than normal predation rates caused by birds and mice). The egg-attacking parasitoids (such as *Fopius arisanus)* have a narrower host range than the larval parasitoids, and have never been recovered from any non-target host in Hawaii.

Experimental results

Several published experimental studies from Hawaii have documented the lack of parasitism of native Hawaiian tephritids by introduced Opiine parasitoids (Duan & Messing 2000 a, b).

In experiments conducted in quarantine over the past several years, *F. ceratitivorus* has been shown to pose no risk to native Hawaiian tephritids that are flower head feeders (Wang et al. 2004) nor to exotic gall formers (Bokonon-Ganta et al. 2005, Bokonon-Ganta & Messing, *in review*).

Other researchers have also studied the host specificity of egg-attacking opine parasitoids. In California it was shown that "*laboratory studies with F. arisanus indicate that it is a specialist on fruit-feeding flies, and will not attack various hosts that feed in flower bids or galls*" (Picket et al. 2008).

Thus theoretical, empirical, and experimental evidence all indicate that there is extremely limited potential for *F. ceratitivorus to attack non-targets in Hawaii.*

V. List of Agencies and Persons Consulted

Dr. Robert Wharton (world expert on Braconidae; Opiinae)
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VI. List of Required Permits and Approvals

Anticipated permits and approvals are listed below:

USDA-APHIS-PPQ (Permit No. 69250 of November 30, 2004)

HDOA (Importation Permit, Pending)

VI. References

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VIII. Appendices