RELEASE OF EXOTIC ORGANISMS: INFORMATION REQUESTED FROM APPLICANTS IN SUPPORT OF PERMIT APPLICATIONS

1. Brief Description of the Proposed Action

The goal is to release a parasitoid, *Lipolexis scutellaris* Mackauer (Hymenoptera: Aphidiidae), of the Brown Citrus Aphid (BCA) in Florida. Petr Stary (Personal communication, May 28/1999) indicated that the name of this species will be changed soon because "...we have to change this name later for [sic] *Lipolexis oregmae* (Gahan) because of priority reasons (this is to be suggested in one of our papers on Guam)" (Appendix 3). However, until the name has been changed formally, we will call this parasitoid *L. scutellaris* (Guam biotype).

The BCA was first detected in Florida in November of 1995 in Dade and Broward counties (Halbert and Brown 1996). The BCA now has established throughout the citrus growing region of Florida. The establishment of *L. scutellaris* could reduce BCA populations in groves and dooryard situations and may slow transmission of severe isolates of citrus tristeza virus (CTV), a serious disease of citrus.

Adults of *L. scutellaris* were imported into quarantine from Guam under USDA-APHIS Permit 954945 (Appendix 1). Live adults of *L. scutellaris* were received in the high security quarantine at the Division of Plant Industry (DPI), Gainesville, Florida on August 19,1999**.** On December 6, 1999 a subculture was transferred, with permission (DACS Form 08208, Appendix 2) to the Department of Entomology and Nematology quarantine facility at the University of Florida, Gainesville. The parasitoids were confirmed as *L. scutellaris* by P. Stary (Appendix 3).

2. Detailed Description of the Proposed Action

• The purpose of the release is to obtain permanent establishment of *L. scutellaris* on *T. citricida* in Florida.

• The *L. scutellaris* colony currently is reared in quarantine facilities in the DPI and in the Department of Entomology and Nematology, University of Florida, Gainesville (UF). If approval is granted to release *L. scutellaris* into the environment, *L. scutellaris* will be reared, out of quarantine, in cages held in greenhouses and screenhouses in Gainesville. We have adequate space and resources to produce approximately 2000 to 3000 parasitoids per month during the remainder of the 2000 growing season. We are able to rear the BCA outside of quarantine because this pest is well established in Alachua County.

• *L. scutellaris* will be released throughout the citrus-growing area in Florida for up to three years, depending on the rate of establishment and spread of this natural enemy. Releases of a <u>minimum</u> of 100 to 200 adults per release site are planned, so we could, potentially, release at 20 to 30 different sites each month. Citrus groves will serve as release sites if the owner agrees to eliminate toxic pesticides for one year, allow researchers back into the orchard to monitor establishment of the parasitoids, and provide approximately 1 to 2 acres for this purpose so that a "buffer zone" is present to reduce potential mortality by toxic pesticide spray drift in adjacent citrus trees. Based on information obtained regarding toxicity of pesticides to the citrus leafminer parasitoid *Ageniaspis citricola*, we expect that citrus growers could safely apply oil and copper for pest and disease control in the release sites (Villanueva-Jimenez and Hoy 1998a, b). The most favorable release sites in citrus groves will be where trees of mixed ages are present so that flushes are frequent, which helps to maintain aphid populations in the grove.

Release sites will be selected by us and by future collaborators, likely including Dr. Jorge Pena (UF-Institute of Food and Agricultural Sciences, Homestead), Dr. Phil Stansly (UF-IFAS, Immokalee), and Dr. J.P. Michaud (UF-IFAS, Lake Alfred), and Dr. David Hall, U.S. Sugar), as well as DPI staff. In addition, Ru Nguyen and Marjorie Hoy will make releases in additional sites, as appropriate. Once releases are made, establishment, overwintering, and dispersal of the parasitoids will be monitored periodically by us and collaborators.

3. Biology of target (host) organism

• *Toxoptera citricida* was described by Kirkaldy in 1907 but he placed it in the genus *Myzus*. It also has been called *Aphis citricidus, Toxoptera citricidus*, *Paratoxoptera argentiniensis,* and *Aphis tavaresi.* The BCA, or oriental citrus aphid, is a pest of citrus and related Rutaceae in Asia. The BCA frequently is confused with its relative *Toxoptera aurantii* Kirkaldy because these two species have a similar appearance and mixed populations can be found in citrus. This confusion may have led to errors in the record of natural enemy species found on the BCA.

The BCA is found in most parts of southern Asia, where it is considered a major citrus pest. The BCA has moved out of Asia and now is found in Africa south of the Sahara, Australia, New Zealand, the Pacific Islands, South America, and the Caribbean. The BCA apparently is able to tolerate a range of physical conditions.

The BCA apparently prefers citrus species and a few closely-related Rutaceae. It has been recorded from other host plants, but it is possible that these records are misidentifications or due to the aphids resting on the plants without feeding or developing (Halbert and Brown 1996). The BCA may be genetically variable and thus it is possible that different 'biotypes' exist, which could have different host plant ranges and different natural enemies.

The BCA has a relatively simple life history. There is no sexual cycle in the fall, as there is with many aphid species, and thus no males, no oviparae (females that deposit fertilized eggs), and no eggs. All individuals are parthenogenetic, producing live young. A single female can initiate a colony, and populations of the BCA can increase very rapidly. Nymphs mature in six to eight days at temperatures of 20^{o}C or higher, with a single aphid able to produce a population of 4,400 within three weeks if natural enemies are absent (Halbert and Brown 1996).

Control of the BCA. By the time the BCA was detected in Florida, it had spread through a sufficiently large area that it was not considered amenable to eradication efforts. In the future, the BCA could colonize other citrus growing regions in the USA, including Louisiana, Texas, Arizona, and California.

Pesticides are applied frequently in Asia to suppress BCA populations and to decrease the rate of spread of CTV disease. Many of these pesticides are toxic to natural enemies. Sprays to control the BCA in Florida could disrupt the effective biological control of other citrus pests. Our citrus IPM program in Florida is very strongly based on biological control, with scale insects, whiteflies, blackflies, mealybugs, citrus leafminer, and mites under substantial to complete biological control.

Citrus propagated on sour orange rootstocks are susceptible to CTV. Yokomi et al. (1993) estimated that at least 18 million citrus trees in Florida are on CTV-sensitive sour orange rootstock. Thus, replanting of substantial acreage will be necessary in the near future. In addition, new budwood must be maintained free of CTV because CTV is graft-transmissible.

Indigenous natural enemies of the BCA in Florida include parasitoids, predators, and pathogens, especially fungi. It is possible that native fungi will assist in the suppression of the BCA in Florida during the summer rainy season. Evans and Stange (1997) surveyed BCA populations in Florida and found several coccinellid predators and two parasitoid species. The most common parasitoid was *Lysiphlebus testaceipes* (Cresson). *Lysiphlebus testaceipes* attacks and kills the BCA but rarely produces viable adults in the BCA in Florida, so the BCA serves as a 'sink'. In addition, *Aphelinus gossypii* Timberlake, which was introduced into Florida from Hong Kong in 1963 for control of *Aphis spiraecola*, has been recovered from the BCA but is more commonly found attacking other aphid species.

Four species of hyperparasitoids, *Alloxysta megourae* complex, *Ceraphron* sp., *Syrphophagus aphidivorus* (Mayr) and *Pachyneuron aphidis* (Bouche), were found parasitizing *L. testaceipes* larvae within the BCA in Florida (Evans and Stange 1997). *Alloxysta pleuralis* was found attacking *L. scutellaris* in India (Singh and Srivastava 1990), so it is possible that *A. megourae* complex hyperparasitoids already present in Florida could attack *L. scutellaris* if *L. scutellaris* is successfully established in Florida.

Yokomi et al. (1993) considered the BCA could become such a serious pest in Florida that they proposed introducing parasitoids into Florida in advance of its arrival in the state, in a tactic they termed "preemptive biocontrol". The goal was to establish species such as *Aphelinus* nr. *gossypii* on alternative host aphids prior to the invasion of the BCA.

Relatively few species of parasitoids are associated with the BCA in the Asian-Pacific region (P. Stary, personal communication, Yokomi et al. 1993, Michaud, 1998). An exception is *Lysiphlebia japonica* (Ashmead), from Japan. *L. japonica* was imported into Florida and Puerto Rico in 1996 and released but, as of 1997, there was no evidence that it had established (Michaud 1998, Evans and Stange 1997). *Lysiphlebia* *mirzai* Shuja-Uddin was imported into Florida from China in 1996 but was not released and the colony is no longer available. *Aphidius colemani* Viereck was imported from Chile but, as of 1997, had not been released in Florida (Evans and Stange 1997).

Four aphid species other than *T. citricida* an be found on citrus in Florida, including *Aphis craccivora* Koch (cowpea aphid), *Aphis gossypii* Clover (cotton or melon aphid), *Aphis spiraecola* Patch (spirea aphid) and *Toxoptera aurantii* (black citrus aphid). All four aphids are reported to be suitable hosts for *L. scutellaris* in the literature (see below).

• The BCA causes economic losses both in groves and nurseries by direct feeding and by efficiently transmitting citrus tristeza virus (CTV) (Komazaki 1994).

Adults and nymphs of the BCA feed on young citrus foliage, depleting the sap. Their feeding can destroy the growing tip of citrus shoots. They also produce honeydew, which allows the growth of sooty mold. More importantly, this aphid is able to transmit severe isolates of CTV more efficiently than other aphids found on citrus in Florida, such as *Aphis gossypii* (Michaud 1998; Yokomi et al. 1989,1994; Rochapena et al. 1995).

Detailed knowledge of the mechanisms by which *T. citricida* transmits CTV is lacking (Halbert and Brown 1996) but CTV is a phloem-limited virus with mild and severe strains. CTV is transmitted in a non-persistent manner by the BCA, meaning that if the aphid is removed from a CTV-infected plant, it can no longer transmit the virus after ca. 48 hours. CTV does not multiply in the aphid. CTV also is transmitted by mechanical inoculation and grafting, but not by seed.

Powell et al. (1997) compared the effectiveness of five aphid control regimes in delaying movement of mild and severe isolates of CTV into a CTV-free sweet orange scion on sour orange rootstock block in Florida over a five-year interval. Trees were treated with Temik, Temik + Meta-Systox R, Meta-Systox R, oil or nothing, respectively. At the end of five years the percentage of trees infected with CTV was not different among the treatments, indicating that pesticides by themselves are not very effective in reducing spread of severe isolates of CTV.

4. Biology of the parasitoid to be released: *Lipolexis scutellaris* (Hymenoptera: Aphidiidae)

• Biology of the Aphidiidae

The Aphidiidae are small hymenopterans consisting of approximately 60 genera and subgenera and more than 400 species from around the world (Stary 1988). *Lipolexis* is one of the most common genera.

The endoparasitic Aphidiidae usually have four larval instars (Stary 1988). Before completing its development, the larva spins a cocoon inside or under the empty aphid exoskeleton. At this stage, the aphid exoskeleton becomes a 'mummy'. The prepupal, pupal and adult stages develop within the cocoon within the mummy. The adult emergence hole, which is circular, is easily broken. The newly emerged adults need a short time to mature. Males often emerge a bit earlier than females and mating occurs soon, lasting several seconds. Aphidiids typically are arrhenotokous, with mated females producing fertilized (diploid) eggs that develop into females and unmated females producing unfertilized (haploid) eggs that develop into males.

The aphidiid developmental rate is influenced by temperature but usually takes two weeks. Adults are active on warm sunny days, especially in late morning and afternoon, showing a positive phototactic response. Longevity is variable; minimum survival occurs without water and food. The sex ratio in the field typically favors females but is variable due to environmental factors. Oviposition may occur soon after females emerge. Females mate only once, but males can mate several times. Oviposition behavior typically involves antennal tapping of the aphid host, then the female stands on erect legs, bending her abdomen forwards beneath the thorax and between her legs. By moving the abdomen forward, she stings the aphid with her ovipositor. The duration of oviposition ranges from about one second to about one minute. The ovipositing female typically can discriminate between aphids that are previously parasitized, thus avoiding superparasitism.

Host instar preference by the female may vary among species within the Aphidiidae. The reproductive capacity also is variable, with up to several hundreds of eggs per female (Stary 1988). Aphidiids disperse in different ways; adults can disperse by flight or by walking, or immatures can disperse within their live aphid hosts or within mummies. Differences in short-distance dispersal can be found between parasitized live aphids and unparasitized aphids (Stary 1988). Prior to mummification, parasitized aphids typically leave their colony and move to microhabitats that are microclimatically favorable for the parasitoids. Long-distance dispersal of adult parasitoids and parasitized alate aphids occurs by flight (Stary 1988).

The Aphidiidae are parasitoids only of the Aphidoidea (aphids) (Stary 1988). Within the family, some parasitoids are restricted to a single host species (monophagy), to two or more species of the same aphid genus, to two or more genera of the same aphid subfamily, to two or more genera of two or more subfamilies of the same aphid family, or to two or more aphid families (Stary 1988). Host finding starts with the selection of a suitable habitat, with the food plants of the host aphids playing an important role, because the parasitoids seem to be attracted to them (Stary 1988). "The attractiveness of the host aphids to the parasitoids is apparently due to the perception of their kairomones, which seem to be present in honeydew. Host and parasitoid population densities are also important in conditioning the searching activity of the parasitoid" (Stary 1988).

Although parasitism ultimately results in mummification and death, parasitized aphids can develop for some time (Stary 1988). If the first or second instars are parasitized, the aphids do not develop to adults. Aphids parasitized in instar 3 reach the adult stage, producing few or no offspring prior to mummification. Aphids parasitized in instar 4 or as adults do reproduce but to a limited extent. Parasitized aphids consume more food but assimilate it less efficiently. They also gain more weight than unparasitized aphids. Parasitized aphids also produce more honeydew.

• Natural enemies of aphidiid parasitoids

Aphidiids are hyperparasitized by Chalcididae, Aphelinidae, Ceraphronidae and Cynipidae (Stary 1988). Predators of aphids may not distinguish between parasitized or unparasitized aphids. Fungi are able to infect both parasitized and unparasitized aphids. Ants often attend aphids, yet some aphidiid species can parasitize the ant-attended aphids.

• Genetic variability within aphidiid parasitoids

Stary (1988) indicated that genetic variability within species of aphidiids is substantial, suggesting that some species actually may consist of species complexes. This type of genetic variability could make it inappropriate to compare the biology of *L. scutellaris* from Guam with that of *L. scutellaris* from India. For example, Stary (1988) pointed out that aphidiids from the same host species but from different areas of the range may differ in developmental rate, host species preference, and adaptation to new climatic conditions. Partial reproductive incompatibility may occur between the different aphidiid populations. Within a defined geographical area, populations on different host species show various genetic diversity patterns. Stary (1988) concluded, "It is recommended to classify all parasitoid populations derived from the same host species in a defined area as a parasitoid biotype. A certain number of strains can be distinguished within this biotype." As a result of this advice, we are calling our population of *L. scutellaris* the 'Guam biotype', even though Dr. Stary suspects it was accidentally introduced into Guam and is not native there (see below).

L. scutellaris is a koinobiont. As noted by Godfray (1994), koinobionts some advantages as natural enemies. Koinobionts can delay their development until the host has matured, which leads to relatively large adult parasitoids that may be "...able to locate more hosts. On the other hand, the specialist adaptations required by exposure to host defenses in endoparasitism result in a narrow host range and tend to reduce the number of hosts encountered. This trend may be offset by the greater host synchronization and specialism that a narrow host range allows..." (Godfray 1994).

• Guam biotype of *L. scutellaris*

P. Stary (Personal communication, Oct. 15, 1999) suspects that *L. scutellaris* "...is obviously an accidental immigrant to Guam, presumably from the Philippines as a part of its natural range. The date of its introduction is unknown as there are no earlier data on the aphidiines in Guam."

We obtained adults of *L. scutellaris* from Guam courtesy of Dr. Ross Miller in August 1999. *L. scutellaris* is a primary parasitoid of the BCA, but detailed reports on its biology in Guam are lacking. Dr. Ross Miller kindly provided some information

(Personal communication, Appendix 5) about the efficacy of *L. scutellaris* as a parasitoid of the BCA in Guam. He noted that

> "*Lipolexis scutellaris* was likely introduced to Guam from the Asian mainland 15-20 years ago in the form of mummies of citrus aphids on introduced citrus plants. Since then it has established itself as the major aphidiid parasitoid of crop aphids on Guam. Its host range on Guam consists of *Toxoptera citricida, Aphis gossypii, Aphis craccivora* and *Aphis spiraecola*. It is most commonly found, and found in the greatest densities, on *T. citricida* on tangerine, lemon, calamondin, and orange, and *A. gossypii* on cucumber, eggplant, melon and squash. Both of these aphids are serious aphid pests of Guam, with *A. gossypii* having the widest host range. Our data show that *L. scutellaris* is specific to aphids on Guam as are other aphidiids. We feel that *L. scutellaris* is an excellent biocontrol agent with moderate dispersal abilities typical of micro-hymenopterans. We have observed infestation rates as high as 90% on *A. gossypii* on cucumber in small (< 5 ha) farms. *L. scutellaris* appears to be quite resilient to catastrophic disturbances of its habitat. It was abundant throughout its former range within 6 months of Supertyphoon Paka, which hit Guam in December 1997. It does, however, appear quite susceptible to chemical sprays used on some farmer fields."

On May 12,1999, Ross Miller (personal communication) indicated that parasitoids "...appear most numerous during the rainy season, which extends roughly from July through December", suggesting this biotype/species requires high relative humidities as adults.

L. scutellaris kills aphids by parasitism. Observations in the Gainesville quarantine indicate that females prefer to oviposit within small aphids. *L. scutellaris* has a relatively short generation time (ca. 12-14 days at ca. 75-78^{o}F in our quarantine facility).

In Guam, *L. scutellaris* has been described as an abundant and effective parasitoid of the BCA (R. Miller, personal communication). However, its effectiveness in suppressing BCA populations under Florida field conditions will have to be analyzed. Because *L. scutellaris* mummies are found in the soil under the citrus tree when reared in pots in quarantine (R. Nguyen, M. Hoy and S. Hill, personal observations), it will be interesting to determine whether the imported red fire ant attacks and kills mummies of *L. scutellaris* in Florida's citrus groves if that behavior is observed in the field. On the other hand, some other ant species have been observed defending aphid mummies, which could enhance the likelihood the parasitoid is able to emerge (Stary 1988). "Reports of direct attack and destruction of mummies by ants are rare and are restricted to some special cases" (Stary 1988).

• Indian biotype of *L. scutellaris*

Singh and Pandey (1997) reported that only mated females of *L. scutellaris* in India oviposit. According to Radhakrishnan and Muraleedharan (1992), when *L.*

scutellaris is reared on *Toxoptera aurantii* in India, the preoviposition period is 3.1 hours, the oviposition period is 2 days, the number of eggs laid per female averages 121, the incubation period averages 3.2 days, the duration of larval instars averages 6.9 days, and the pupal period is 5.8 days. Adult females live an average of 5.1 days, while males live only 2.6 days. Radhakrishnan and Muraleedharan (1992) indicated that "Parasitised aphids were generally avoided" by *L. scutellaris* in India and "All the three aphidiid species showed a high degree of discrimination in selecting the specific instar of the host. Mostly, second and rarely third instar nymphs of *T. aurantii* were selected for egg laying." Biswas and Singh (1995) evaluated the number of males and females of *L. scutellaris* in field populations in India. They found that the proportion of males ranged from 29 to 37%.

Dharmadhikari and Ramaseshiah (1970) reported on the biology of *L. scutellaris* in India. According to them, *L. scutellaris* "...prefers the host genera, *Aphis* and *Toxoptera*." They report the parasitoid's effectiveness was reduced by hyperparasitoids in the genera *Alloxysta* and *Aphidencyrtus.* The life cycle from egg to adult emergence was 12.5 days on average during August-September, with mummies starting to appear on the 5th day after oviposition. With the onset of winter, this period was lengthened by 3 to 4 days, adults emerging after 8 to 9 days. Dharmadhikari and Rameseshiah (1970) reported the mummies are "...pale pinkish to brick red with cottony white cocoons appearing through slits in the bodies." Diapausing and non-diapausing mummies were described by Shuja-Uddin (1977).

This description of the Asian biotype mummy is slightly different from what we have seen in our rearing of *L. scutellaris* from Guam on the BCA. It is possible that this is due to the population in India being reared on a different aphid species *(T. aurantii)* or to a different 'biotype' of *L. scutellaris.* Dharmadhikari and Ramaseshiah (1970) reported *L. scutellaris* "...adults lived up to 5 days in field cages which had been prepared in the laboratory premises. Adults lived 1-2 days longer when nectar-bearing flowers of *Impatiens balsamina* were provided in these cages." They further reported that "The females accept almost any stage of the host aphid for oviposition."

Host Range of *L. scutellaris*

Information on the host range of *L. scutellaris* is based on taxonomic records of material collected from field samples. There surely are errors in identification of both host aphids and parasitoids, but the data do serve as an indicator of potential host range. *L. scutellaris* is not known to be a hyperparasitoid.

In India, *L. scutellaris* is recorded on the following aphid species: *Aphis citricola*, *Aphis craccivora, Aphis fabae, Aphis gossypii, Aphis nerii, Aphis ruborum longisetosus, Aphis solanella, Toxoptera aurantii, Toxoptera odinae* (Stary and Ghosh 1983, Ahmad and Singh 1996). Radhakrishnan and Muraleedharan (1992) reported that *L. scutellaris* was a parasitoid of *Toxoptera aurantii* on tea in south India. Singh and Tripathi (1987) recorded *L. scutellaris* from four aphid species in India: *Aphis gossypii, A. craccivora, Myzus avenae,* and *M. persicae*. Singh and Srivastava (1990) reported collecting *L. scutellaris* from the following aphids in India: *Aphis citricola, A. craccivora, A. fabae, A.*

gossypii, A. nerii, A. ruborum longisetosus, Trichosiphum formosana, Myzus persicae, Macrosiphum avenae, Rhopalosiphum maidis, R. nympheae, Toxoptera aurantii, T. citricidus, T. odinae, and *Tuberolachnus salignus*.

In Taiwan, *L. scutellaris* was reported parasitizing *Aphis laburni* Kalt. on beans; and parasitizing *A. spiraecola*, *Toxoptera aurantii* and *T. citricidus* on *Citrus* sp. (Chiu and Liu 1969).

In Vietnam, Stary and Zeleny (1983) reported finding *L. scutellaris* on *Aphis citricola* (= BCA), *Aphis gossypii, A. nerii,* and *Rhopalosiphum nympheae.* They reported the mummies as "...light yellowish to whitish, not directly attached to the leaf. No diapause mummies were found." Stary and Zeleny (1983) reported that *L. scutellaris* is a widely distributed species in India, Pakistan, southern China, Taiwan, Vietnam, and Japan.

Stary and Zeleny (1983) evaluated published records and host records in Vietnam and concluded that "...*Lipolexis scutellaris* is a relatively broadly oligophagous species on *Aphis, Rhopalosiphum, Toxoptera* and other aphids." They further concluded that "*Lipolexis scutellaris* has a promising host range from the biological control viewpoint: *Aphis citricola, Aphis gossypii, Aphis craccivora* and others are widely distributed pests in the tropics and more or less also in the other zones."

• Potential effects on weed biological control programs

Dr. Jim Cuda (personal communication) indicated that he knows of only one aphid imported into North America as a weed biological control agent. The aphid is *Aphis chloris* Koch, which was imported in 1979 into Canada (British Columbia and Nova Scotia) from Germany as a natural enemy of *Hypericum perforatum* Linnaeus (Julien and Griffiths 1998). It seems unlikely that *L. scutellaris* would disperse to Canada or be climatically adapted to survive and establish in Canada. Thus, release of *L. scutellaris* is unlikely to interfere with this weed control program.

Dr. Cuda indicated he did not know of any other weed control program in which an aphid is under consideration as a natural enemy in the USA.

• Potential effects of *L. scutellaris* on rare endemic aphids

Dr. Susan Halbert, an expert on aphids at the Division of Plant Industry, Florida Department of Agriculture and Consumer Services, discussed whether *L. scutellaris* "...might attack nontarget aphids and if so, whether the environmental consequences would be serious enough to abandon plans for release. Any deliberate release of an exotic organism requires thoughtful consideration, but in the end, the decision must be made based on weighing the risks and the benefits" (Appendix 6).

Dr. Halbert noted that most aphid parasitoids are not particularly specific. Thus, Dr. Halbert addressed two questions: "...are there rare and endangered aphids in

Florida that may be impacted, and second, how could this be assessed in the laboratory?" Dr. Halbert indicated there are rare indigenous aphids in Florida, including *Aphis minima* (Tissot). She concluded, however, that

> "In my opinion, exotic aphid parasites are not likely to have much negative effect on Florida's ecosystems. Aphids are obligate plant parasites. Without their host plants, they die. In my opinion, the only way to endanger an aphid species is to endanger its host plant. Although non-specific aphid parasites may attack native aphids, they are more likely to attack the relatively abundant pests of commercial and ornamental crops. Aphids are the ultimate r-strategists. Many species are capable of a 1000 fold increase in a 3-week time period. Populations of aphids fluctuate markedly throughout the year under natural conditions and are unlikely to be affected much by a new parasite unless it selectively attacked them at low density. It is improbably that an exotic general parasite would selectively attack an obscure native aphid at low density."

Dr. Halbert indicated that it would be very difficult to experimentally quantify the risk (Appendix 6):

> "First, it would be extremely difficult to obtain and culture all the aphid species required, particularly given that some may not be described, and others haven't been seen in 60 years. Others may require plants that are very difficult to obtain and propagate or are too large to contain in quarantine. Second, results are difficult to interpret. If the parasite absolutely refuses a given species, that is a clear cut case, but it is more likely that parasites will complete development in the laboratory on at least some of the test species. Even if this occurs, it is not clear that parasites will parasitize the same aphids in a natural setting."

Finally, Dr. Halbert concluded (Appendix 6) that there is a strong case for classical biological control of the brown citrus aphid:

> "Brown citrus aphid is an extremely serious pest of citrus. Its introduction into Florida was a tragedy of major proportions. We expect to lose some 20% of our citrus trees (about 20 million trees) in the next few years, as a result of the increase [d] spread of decline strains of citrus tristeza virus (CTV). Those figures are only for Florida. The aphids inevitably will spread to Texas, Arizona and California, where further major losses will occur."

Dr. Halbert goes on to conclude that:

> "In this case, parasites for brown citrus aphid have the potential to help partially mitigate a major agricultural calamity, and the risks to Florida ecosystems are minimal and impossible to demonstrate. In my opinion, the potential benefits to Florida citrus of introducing parasites of brown citrus aphid outweigh the risks."

5. Expected attainable geographic range in North America of the organism to be released

Based on our current knowledge of the biology of the Guam biotype of *L. scutellaris,* its expected geographic range is where the BCA becomes established in citrus. If the BCA spreads to other citrus-growing regions in the USA, *L. scutellaris* could move with the BCA into Louisiana, Texas, Arizona, and California. Because the Guam biotype could have a host range that includes *Aphis gossypii*, *Aphis craccivora* and *Aphis spiraecola* (all of which are on citrus in Florida)*,* the Guam biotype of *L. scutellaris* could be found where these aphids are found, which could include host plants in addition to citrus. Because *L. scutellaris* is from a tropical/subtropical climate, we do not expect it to colonize temperate regions.

6. Expected environmental effects of the proposed release

• *L. scutellaris* is expected to establish on the BCA and to reduce BCA populations. The use of pesticides to control this pest should be reduced, leading to fewer negative effects on a variety of natural enemy species in citrus, or negative effects on the ground water, workers, or consumers.

The following products were recommended as possible chemical control options for the BCA in Florida: Admire and Provado (imidacloprid), dimethoate (Cygon), azinphosmethyl (Guthion), methomyl (Lannate), chlorpyrifos (Lorsban), oxydemeton methyl (Metasystox-R), acephate (Orthene), carbaryl (Sevin), aldicarb (Temik) and endosulfan (Thiodan) (Knapp et al. 1996). Some of these pesticides may become unavailable in the near future if the Environmental Protection Agency recommends that they no longer be registered based on their potential hazards under the Food Quality Protection Act. Cygon, Guthion, Lannate, Lorsban, Orthene, Sevin, and Thiodan are likely to be highly toxic to most species of natural enemies (Villanueva-Jimenez and Hoy 1998b).

It is unlikely that *L. scutellaris* would have significant negative effects on humans or beneficial species of insects such as honeybees. No aphids in Florida have been declared to be threatened or endangered. The potential interactions, under Florida conditions, between *L. scutellaris* and any other parasitoids are difficult to predict.

The role of <u>native</u> <u>hyperparasitoids</u> in reducing the effectiveness of *L. scutellaris* as a natural enemy of BCA in Florida remains to be determined.

• Beneficial effects of the establishment of *L. scutellaris* in Florida could include: reduced populations of BCA in citrus groves and nursery trees, reduced frequency of transmission of CTV in Florida, reduced applications of pesticides to control the BCA, reduced secondary pest outbreaks due to the negative effects of pesticides on other natural enemies of citrus pests. The use of pesticides that exhibit low toxicity to natural enemies is crucial because most citrus pests in Florida are under substantial to complete biological control and applications of pesticides toxic to natural enemies could induce secondary pest outbreaks.

• Prior to release of adult *L. scutellaris,* samples will be preserved as voucher specimens both in 70% (for taxonomic purposes) and in 95% EtOH (for future PCR tests). The specimens preserved in 95% EtOH will frozen at -80^{o}C and stored in the laboratory of Dr. M.A. Hoy. Taxonomic vouchers will be deposited with Dr. Lim Nong (DPI).

CONCLUSIONS

The risks associated with releases of the Guam biotype of *L. scutellaris* appear low while the benefits of such releases could be high.

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