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Field Release of the Gallforming Fly, *Parafreutreta regalis* Munro (Diptera: Tephritidae), for Biological Control of Cape-ivy, *Delairea odorata* (Asterales: Asteraceae), in the Contiguous United States

Environmental Assessment, February 2015 Field Release of the Gall-forming Fly, *Parafreutreta regalis* Munro (Diptera: Tephritidae), for Biological Control of Cape-ivy, *Delairea odorata* (Asterales: Asteraceae), in the Contiguous United States

# Environmental Assessment, February 2015

#### **Agency Contact:**

Shirley Wager-Page, Branch Chief Regulations, Permits, and Manuals Plant Protection and Quarantine Animal and Plant Health Inspection Service U.S. Department of Agriculture 4700 River Road, Unit 133 Riverdale, MD 20737–1236

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# I. Purpose and Need for the Proposed Action

The U.S. Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS), Plant Protection and Quarantine (PPQ), Pest Permitting Branch (PPB) is proposing to issue permits for release of a gall-forming fly, *Parafreutreta regalis* Munro (Diptera: Tephritidae). The agent would be used by the applicant for the biological control of Capeivy, *Delairea odorata* (Asterales: Asteraceae), in the contiguous United States.

This environmental assessment<sup>1</sup> (EA) has been prepared, consistent with USDA, APHIS' National Environmental Policy Act of 1969 (NEPA) implementing procedures (Title 7 of the Code of Federal Regulations (CFR), part 372). It examines the potential effects on the quality of the human environment that may be associated with the release of *P. regalis* to control infestations of Cape-ivy within the contiguous United States. This EA considers the potential effects of the proposed action and its alternatives, including no action.

APHIS has the authority to regulate biological control organisms under the Plant Protection Act of 2000 (Title IV of Pub. L. 106–224). Applicants who wish to study and release biological control organisms into the United States must receive PPQ Form 526 permits for such activities. The PPB received a permit application requesting environmental release of a gall forming fly, *P. regalis*, from South Africa, and the PPB is proposing to issue permits for this action. Before permits are issued, the PPB must analyze the potential impacts of the release of this agent into the continental United States.

The applicant's purpose for releasing *P. regalis* is to reduce the severity of infestations of invasive Cape-ivy in the continental United States. A native of South Africa, Cape-ivy has recently become one of the most pervasive non-native plants to invade the coastal areas of the western United States, spreading in riparian (living on the bank of a natural watercourse, lake or tidewater) forests, coastal scrubland, grassland, Monterey pine forest, coastal bluff communities, and seasonal wetlands, particularly in California and Oregon. Though this weedy vine prefers moist, partly-shaded environments along the Pacific coast, there are increasing reports of infestations at inland riparian locations. The vine was imported as an ornamental in North America, and this is the probable

<sup>&</sup>lt;sup>1</sup> Regulations implementing the National Environmental Policy Act of 1969 (42 United States Code 4321 et seq.) provide that an environmental assessment "shall include brief discussions of the need for the proposal, of alternatives as required by section 102(2)(E), of the environmental impacts of the proposed action and alternatives, and a listing of agencies and persons consulted." 40 CFR § 1508.9.

source of introduction and spread. Fragments of the plant easily root and this characteristic has facilitated its spread. The climbing vine causes serious environmental problems by overgrowing riparian and coastal vegetation, causing other plants to die. Along California's coast, it is also a roadside weed that requires frequent herbicide treatment.

Existing management options for management of Cape-ivy are expensive, temporary, and have nontarget impacts. For these reasons, the applicant has a need for environmental release of *P. regalis*, a host-specific, biological control organism for the control of Cape-ivy. Galling of Cape-ivy caused by *P. regalis* greatly reduces growth of infested plants, which should reduce this vine's negative impacts.

# II. Alternatives

This section will explain the two alternatives available to the PPB—no action and issuance of permits for environmental release of *P. regalis*. Although the PPB's alternatives are limited to a decision on whether to issue permits for release of *P. regalis*, other methods available for control of Cape-ivy are also described. These control methods are not decisions to be made by the PPB, and their use is likely to continue whether or not the PPB issues permits for environmental release of *P. regalis*, depending on the efficacy of *P. regalis* to control Cape-ivy. These are methods presently being used to control Cape-ivy by public and private concerns.

A third alternative was considered, but will not be analyzed further. Under this third alternative, the PPB would have issued permits for the field release of *P. regalis*; however, the permits would contain special provisions or requirements concerning release procedures or mitigating measures. No issues have been raised that would indicate special provisions or requirements are necessary.

#### A. No Action

Under the no action alternative, the PPB would not issue permits for the field release of *P. regalis* for the control of Cape-ivy. The release of this biological control agent would not take place. The following methods are presently being used to control Cape-ivy; these methods will continue under the "No Action" alternative and will likely continue even if the PPB does not issue permits for release of *P. regalis*, depending on the efficacy of the organism to control Cape-ivy.

1. Chemical<br/>ControlSeveral herbicides, such as glyphosate (Roundup®) or clopyralid<br/>(Transline®) when used with appropriate surfactants, can provide<br/>temporary control of Cape-ivy. Cape-ivy now infests many natural areas

where some control measures such as herbicides are restricted or even prohibited.

- 2. Mechanical Control Mechanical control of Cape-ivy has had limited success but is usually manually removed by ripping the vines out. In Golden Gate National Recreation Area, Cape-ivy is mainly removed manually or with power tools, but other methods such as goat grazing and prescribed fire are being used.
- **3. Biological Control** Another biological control agent for Cape-ivy is proposed for environmental release. The agent is a stem-boring moth, *Digitivalva delaireae*, also from South Africa. The agent would also be used by the permit applicant for biological control of Cape-ivy.

# B. Issue Permits for Environmental Release of *P. regalis.*

Under this alternative, the PPB would issue permits for the field release of the gall-forming fly, *P. regalis*, for the control of Cape-ivy. These permits would contain no special provisions or requirements concerning release procedures or mitigating measures.

#### **Biological Control Organism Information**

**1. Taxonomy**Common name: Cape-ivy gall fly (unofficial)<br/>Scientific name: Parafreutreta regalis Munro (Diptera: Tephritidae)

Classification from "The Diptera Site" (Norrbom et al., 1999)

Phylum: Arthropoda Class: Insecta Order: Diptera Suborder: Brachycera Infraorder: Muscomorpha Family: Tephritidae Subfamily: Tephritinae Tribe: Tephritini Genus group: Sphenella Genus: *Parafreutreta* Species: *P. regalis* 

#### a. Native Range

2. Geographical Range of P. regalis P. regalis was originally described from a single collection in what is now called Kwazulu-Natal Province (Munro, 1940). During two years of surveys in South Africa this fly was shown to be widespread through most of the range of Cape-ivy, but never very abundant (Grobbelaar, 1999; Grobbelaar et al., 2003). This region is between 25 and 33.5° S latitude (USDA, ARS, 2012).

#### b. Expected Attainable Range of P. regalis in North America

The applicants recovered *P. regalis* from galls collected at a variety of sites where Cape-ivy occurs in South Africa, including mountain ranges and coastal sites; thus, *P. regalis* should be climatically suited to establish at all Cape-ivy sites in California and southwestern Oregon. (USDA, ARS, 2012).

#### P. regalis adult females emerging from galls soon mate. Galls are growths 3. Life History of that develop on Cape-ivy in reaction to the feeding of *P. regalis* inside the P. regalis stem. Within 24 hours of mating, females begin inserting eggs into the stem nodes of Cape-ivy vines (Balciunas and Mehelis, 2010). A node is the area of a plant's stem from where the leaves grow. *P. regalis* eggs are opaque, longer than they are wide, and about 0.5 millimeters (mm) long. The female fly inserts a group of 3 to 28 eggs into a Cape-ivy node. Females lay (oviposit) an average of 61 eggs during their lifetime. After 7 days, the eggs begin to hatch, and a few days later, the first swelling of the nodes become visible, indicating the beginning of gall formation. P. regalis undergoes three larval instars (worm-like stages of an immature insect), each lasting approximately a week. Before pupating inside the gall, the late third instar larva chews an exit hole in the interior of the gall, that is covered by a 'window' of intact plant cuticle. About 20 days after the window appears, the adult insect emerges from the pupal case inside the gall, it breaks through the cuticle covering the window, and it escapes from the plant to mate. The time from egg to adult takes about two months (mean = 56 days). Adults usually live about two weeks, but some may live up to four weeks. Under laboratory conditions, six generations per year are possible. (From USDA, ARS, 2012)

## **III. Affected Environment**

#### A. Target Weed

Common names include: Cape-ivy, Cape ivy, German ivy, parlor ivy and Italian ivy.

Phylum: Magnoliophyta Class: Magnoliopsida Order: Asterales Family: Asteraceae Subfamily: Asteroideae Tribe: Senecioneae Subtribe: Senecioninae Genus: *Delairea* Species: *D. odorata* Lemaire

Cape-ivy is a perennial, slender stemmed, creeping and twining vine that can climb up to 9 meters (approximately 30 feet) (DiTomaso and Healy, 2007). It grows in deep shade or cloudy conditions, and does not usually tolerate full sunlight. Vines are evergreen in mild climates, but aerial parts of the plant are susceptible to frost. The plant has rhizomes (underground plant stems capable of producing the shoot and root systems of a new plant), stolons (a stem, at or just below the surface of the ground, that produces new plants from buds at its tips or nodes), and short fibrous roots. Stems of Cape-ivy usually have flattened, ear-like stipules or outgrowths at most nodes of the plant. Cape-ivy plants that have stipules are considered "stipulate" and Cape-ivy plants without stipules are considered "exstipulate". In South Africa, generally only the stipulate variety is found, with the exception of one known population that is exstipulate. In California, both stipulate and exstipulate varieties are common (Robison and DiTomaso, 2010).

#### B. Areas Affected by Cape-ivy

#### 1. Native and Introduced Range of Cape-ivy

The native range of Cape-ivy is South Africa. Despite the restricted habitat and geographic range in its native South Africa, Cape-ivy has shown an ability to adapt to an array of habitats when introduced into foreign countries, and at some locations, to expand its environmental range. In the Southern Hemisphere, it has become naturalized in Argentina (Parodi, 1959; Hilliard, 1977), Australia (Lamp and Collet, 1979; Fagg, 1989; Blood, 2001), and New Zealand (Webb et al., 1988; Owen, 1996). It is considered a problematic weed in the latter two countries.

In Europe, it has escaped from cultivation in parts of the Low Countries and on the south coast of England (Hilliard, 1977). Cape-ivy is naturalized in at least three regions of Italy (Catalano et al., 1996), and it is becoming a problem in northern Spain (de la Torre-Fernández and Álvarez-Arbesú, 1999), and Portugal (Palhinha, 1974). A recent "Compendium" on crops and weeds, also adds three islands: Azores, Corsica, and St. Helena to the countries listed above (CABI, 2011).

The earliest known specimen from the United States was collected in Berkeley, California in 1892 (Robison and DiTomaso, 2010). Jepson (1925) recorded it as naturalized, but listed only four locations. Munz (1959) listed this vine as naturalized in canyons and gullies along the coast of south and central California. Barkley (1993) indicated that it was "highly invasive" and added the northern coastal regions to the areas with known infestations. A 1999 survey effort (Robison et al., 2000; Robison and DiTomaso, 2010) not only confirmed that it was widespread along most of coastal California, but added some inland sites, as well as an infestation in southern Oregon. Cape-ivy occurs throughout all coastal counties of California, as well as the Channel Islands (Santa Rosa, Santa Cruz, and Santa Catalina) and Curry County, Oregon. Geographic Information System-based climatic analysis, based on its occurrence in California, indicates that Cape-ivy occurs at elevations between 0 and 891 meters (2,923 feet), annual mean temperatures between 10.5 and 17.7° C, and in areas with annual precipitation ranging between 232 (9 inches) and 2,270 millimeters (89 inches) (Robison and DiTomaso, 2010). It can tolerate extreme mean monthly temperatures between 1.8 and 31.8°C.

Cape-ivy was introduced to the Big Island of Hawaii around 1909 and has become a serious weed in a variety of upland habitats between 500 (1,640 feet) and 2,500 meters (8,202 feet) in elevation (Jacobi and Warshauer, 1992). Haselwood and Motter (1983), and Jacobi and Warshauer (1992) reported that this vine was restricted to the Big Island in the Hawaiian Islands. However, it has also naturalized on Maui (Wagner et al., 1990), and it is spreading (Starr et al., 2003). It has also been noted on Oahu (HEAR project, 2011). Although Cape-ivy is present in the Hawaiian Islands, *P. regalis* is not proposed for release there at this time.

2. Potential Distribution of Cape-ivy in North America
A rough prediction of the plant's potential geographic distribution was made using the Match Locations component of the CLIMEX computer program (Sutherst and Maywald, 1985). This model describes the response of a species to climate, and enables the user to predict the potential geographical distribution of the species. The Mediterranean template provided a good fit to the known distribution of the plant in South Africa (Figure 1). This model predicts a distribution in the United States that is very similar to that observed by Robison and DiTomaso (2010), restricted to coastal California. Thus the plant may already be close to occupying its potential geographic range.

**Figure 1.** Predicted potential geographic distribution of Cape-ivy based on the Environmental Index calculated using the Mediterranean template in CLIMEX (from USDA, ARS, 2012).



#### C. Plants Related to Cape-ivy and Their Distribution

Plants related taxonomically to Cape-ivy would be the most likely to be attacked by the proposed biological control organism *P. regalis*. Plants related to the target weed Cape-ivy are discussed below.

Cape-ivy belongs to the tribe Senecioneae in the plant family Asteraceae. Senecioneae is the largest tribe in the Asteraceae, comprising about 3,000 species in 150 genera (Bremer, 1994; Pelser et al., 2007). *Delairea odorata* (Cape-ivy) is the only species within the genus *Delairea* (Jeffrey, 1986). *Delairea* is thought to be most closely related to the plant genera *Mikaniopsis*, *Cissampelopsis*, and *Austrosynotis* (Jeffrey, 1986; 1992), none of which are represented in North America. Its closest relatives in North America are the introduced *Erechtites* species (burnweeds) and plants from the genus *Senecio*. The genus *Senecio* 'sensu lato' contains about 1,250 species, but reclassifications in the genus have occurred over the past decades, leading to recognition of new genera (Trock and Barkley, 1998; Barkley, 1999). Thus, some plants formerly classified as *Senecio* species are now recognized as belonging to the genera *Packera* and *Pseudogynoxys*.

*Erechtites* (burnweeds) includes four species in the United States, with two to three occurring in California. *Pseudogynoxys* includes one species that is present in Florida (Mexican flamevine). Both *Senecio* and *Packera* genera are large and diverse, with 30 species, varieties, and subspecies of *Senecios*, and 20 species, varieties, and subspecies of *Packera* in California alone, and 84 *Senecio* and 64 *Packera* species, varieties, and subspecies throughout the United States (USDA, NRCS, 2011). These species are found in a wide range of habitats and locations.

Species that were used in testing the specificity of *P. regalis* to Cape-ivy are listed in appendix 1.

# **IV. Environmental Consequences**

#### A. No Action

#### 1. Impact of Spread of Cape-ivy

Cape-ivy grows rapidly in California, smothering other vegetation on the ground and climbing vertically to a height of eight meters, frequently killing other plant species. In California, Cape-ivy is most invasive in coastal areas, and is especially damaging to riparian forests, but coastal scrub and grassland communities and coastal bluffs are also readily invaded (Balciunas and Archbald, 1999). These areas contain a large share of California's rare and endangered native plants, and Cape-ivy is ranked

in the "List A-1, most invasive" category by California Exotic Pest Plant Council (1994; 1996; 1999). It was considered for listing as a noxious weed by the California Department of Food and Agriculture (CDFA, 2003). In Golden Gate National Recreation Area, an infestation of 3.6 hectares of Cape-ivy in 1987 expanded to 27.2 hectares nine years later (Alvarez, 1998).

A study of three habitats (coastal scrub, willow riparian, and alder riparian) at Golden Gate National Recreation Area found that sites infested by Cape-ivy for five to 10 years had 78 percent fewer annual plant species than uninfested plots, and 10 percent fewer perennial species (Alvarez, 1999). Invaded plots had 31 percent fewer native species and 32 percent fewer non-native species. Overall, invaded plots contained 77 percent fewer seedlings than uninvaded plots (65 percent fewer native vs. 81 percent non-native). Cape-ivy infested plots had 56 percent less bare ground than uninfested plots (2.5 percent vs. 6 percent cover). A two-year study where Cape-ivy was repeatedly removed from the same three habitats resulted in an average increase of plant species by 32 percent, ranging from 24 percent in the willow and alder riparian habitats to 48 percent in the coastal scrub habitat (Alvarez, 1999). Most of the change was attributed to a 400 percent increase in seedling recruitment, primarily of grasses and herbaceous flowering plants, in the absence of Cape-ivy.

An analysis overlaying known and potential locations of Cape-ivy with the location of California sensitive species indicated that 163 sensitive plants are expected to overlap with predicted Cape-ivy sites using a 100-meter buffer around the infested location (Robison and DiTomaso, 2010). Overlap between predicted Cape-ivy infestations and steelhead salmon (*Oncorhynchus mykiss*) populations ranged between 42 and 50 percent. Although no published studies have been reported on the toxicity of Cape-ivy to fish, some evidence (C. Bossard, unpublished data) suggests that Cape-ivy is toxic to the golden shiner (*Notemigonus crysoleucus*). Preliminary experiments exposing mosquito fish (*Gambusia affinis*) to crushed Cape-ivy leaves suggested that toxicity was low (Balciunas, unpublished data).

 2. Impact from Use of Other Control Methods
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 3. The continued use of chemical herbicides, and mechanical and biological control sat current levels would be a result if the "no action" alternative is chosen. These environmental consequences may occur even with the implementation of the biological control alternative, depending on the efficacy of *P. regalis* to reduce Cape-ivy populations in the contiguous United States.

#### a. Chemical Control

The use of herbicides, while effective, is limited to relatively accessible sites. In addition, control is only temporary and other vegetation is

harmed as well (Bossard, 2000). Cape-ivy now infests many natural areas where herbicides are restricted or even prohibited.

#### b. Mechanical Control

Mechanical control is labor intensive and seldom effective because remaining plant parts produce new plants that quickly grow back to fill the void. Because small pieces of the plant can regrow, it is important to ensure that removed plants are not chipped or sent unbagged to a dump site; otherwise, the removed Cape-ivy could be spread to new locations. Mechanically cleared sites require intense monitoring to ensure that new plants do not resprout.

#### c. Biological Control

No organisms for the biological control of Cape-ivy have been released in North America. However, if *P. regalis* is released in combination with the stem-boring moth (*Digitivalva delaireae*), the impact on Cape-ivy control is expected to be even greater than *P. regalis* alone. There is no guarantee that the PPB will issue a permit for *D. delaireae* in the future until additional testing of that insect is completed.

# B. Issue Permits for Environmental Release of *P. regalis*

Impact of P. regalis on Nontarget Plants
 Host specificity of P. regalis to Cape-ivy has been demonstrated through scientific literature, field observations, and host specificity testing. If an insect species only attacks one or a few closely related plant species, the insect is considered to be very host-specific. Host specificity is an essential trait for a biological control organism proposed for environmental release.

#### a. Scientific Literature

*Parafreutreta regalis* has only been recorded from Cape-ivy (Munro, 1940).

#### b. Field Observations

During the two years of surveys conducted by the applicant in South Africa, *P. regalis* was only reared from galls of Cape-ivy (USDA, ARS, 2012). The galls on other species of vines (e.g., *Mikania capensis*, *Senecio deltoideus*, *S. helminthioides*, *S. quinquelobus*, *S. tamoides*) produced a variety of other tephritid species, including *Parafreutreta felina* from *S. tamoides* (Grobbelaar et al., 2003).

#### c. Host Specificity Testing

Host specificity tests are tests to determine how many plant species *P*. *regalis* attacks, and whether nontarget species may be at risk. See appendix 2 for information regarding host specificity testing methods. In host specificity testing, *P. regalis* only formed galls and developed on Cape-ivy. No other plant species were attacked by *P. regalis*, including plant species closely related to Cape-ivy.

#### (1) Site of Quarantine Studies

The individuals of *P. regalis* used in host specificity tests were originally reared from galls collected near the town of Wilderness, in South Africa's Western Cape Province. Hymenopteran parasitoids were excluded from adult insects which were used to establish a laboratory colony. Laboratory studies were conducted at the USDA-ARS quarantine facility in Albany, CA and at the Weeds Programme, Plant Protection Research Institute, in Pretoria, South Africa.

#### (2) Test Plant List

The list of plant species used for host specificity testing of *P. regalis* is shown in appendix 1. The strategy used for selecting plants for testing is based on the phylogenetic approach, where closely related species are theorized to be at greater risk of attack than are distantly related species (Wapshere, 1974).

Plants for testing the host range of *P. regalis* were selected from seven possible categories. Test categories consisted of the following:

CATEGORY 1: Genetic types of Cape-ivy (varieties, races, forms, genotypes, apomicts, etc.).

Both stipulate and exstipulate varieties of Cape-ivy from California were tested (Robison and DiTomaso, 2010).

CATEGORY 2: North American species in the same genus as Cape-ivy.

There are no North American species in the genus Delairea.

CATEGORY 3a: Other genera in the same tribe (Senecioneae) as Cape-Ivy.

Besides Cape-ivy, 46 species/varieties in 15 genera in the tribe Senecioneae were tested by the applicant (USDA, ARS, 2012). The majority of these (41 species in 10 genera) were in the same subtribe, Senecioninae, as Cape-ivy. These included South African vines in the genus *Mikaniopsis* (considered one of the closest relatives to *Delairea*) and *Senecio*. Five species in the other two, much smaller, subtribes, Blennospermatinae and Tussilagininae were also tested.

CATEGORY 3B: Genera in other tribes of Asteraceae.

Delineation of subfamilies within the Asteraceae appears to be in a state of flux. However, tribal classifications appear to be well defined, and currently there is consensus that the family is comprised of 17 tribes (Bremer, 1994; Judd et al., 2002). At least one species in each of the 15 tribes that are represented in the United States was tested, either as native or naturalized species. A total of 38 species/varieties in 35 genera in these other tribes were tested. Crops in the Asteraceae family tested include artichoke, safflower, sunflower, Jerusalem artichoke, chicory, and lettuce.

CATEGORY 4: Threatened and endangered species in the Asteraceae family, divided by subgenus, genus, subfamily, and tribe.

The family Asteraceae is the largest in the world, consisting of more than 1,500 recognized genera and 23,000 known species (Bremer, 1994). Many of these are rare, including approximately 101 species/varieties in the United States that have been federally listed as threatened or endangered (USFWS, 2014). Threatened and endangered plant species in the same tribe (Senecioneae) as Cape-ivy have the most potential for risk from *P. regalis*.

A rare species of *Blennopserma*, *B. bakeri*, is federally listed as endangered. Another variety in this genus, B. nanum var. robustum, is listed by California as rare. A close relative, B. nanum var. namum, was tested, which serves as a surrogate for both species/varieties. Three species of *Packera* are federally or state listed as threatened or rare. Packera ganderi (state-listed in California) was tested as well as three other Packera species. Oregon lists Luina serpentina as threatened, and the only other member of this genus, L. hypoleuca, was tested as a surrogate for it. Oregon also lists Senecio ertterae as threatened. This shrub occurs only in arid far-eastern Oregon, almost 500 kilometers from the closest Cape-ivy infestation. Senecio flaccidus, which occurs in California and is closely related to S. ertterae, and S. aronicoides and S. hydrophilus, which occur in California and Oregon, were tested as surrogates. The federally listed Yermo xanthocephalus occurs only in central Wyoming, and is never likely to be encountered by *P. regalis* because of extreme differences in climate and great distance; thus, this plant was not tested.

CATEGORY 5: North American species in other families in the Asterales order that have some phylogenetic, morphological, or biochemical relationship to Cape-ivy, including economically and environmentally important plants.

Besides the large family Asteraceae, the order Asterales contains a handful of small families, of which Campanulaceae is the largest (Judd et al., 2002). Two species in this related family were tested. Also, four species were tested in the families Amaranthaceae (chard) and Brassicaeae (cabbage, radish), that contain many crop species.

CATEGORY 6: North American species in other orders that have some morphological or biochemical relationship to Cape-ivy, including economically and environmentally important plants.

Seven species of native or naturalized vines in six families were tested: *Hedera canariensis* and *H. helix* (Apiaceae), *Aristolochia californica* (Aristolochiaceae), *Marah fabaceus* (Cucurbitaceae), *Clematis lingusticifolia* (Ranunculaceae), *Fragaria chiloensis* (Rosaceae) and *Vitis californica* (Vitaceae), belonging to six different orders (Judd et al., 2002).

CATEGORY 7: Any plant on which the biological control agent or its close relatives (within the same genus) have been previously recorded to feed and/or reproduce.

All species of *Parafreutreta*, for which hosts are known, attack only *Senecio* spp. or Cape-ivy except for one. The exception is *P. mavoana*, which occurs on Madagascar, and is recorded from *Dodonaea viscosa*, a member of the Sapindaceae family. *Dodonaea viscosa* was tested by the applicant, as well as some other North American representatives of Sapindaceae: two native species of maples (*Acer macrophyllum, Acer negundo* var. *californicum*), western buckeye (*Aesculus californica*) and balloon vine (*Cladiospermum halicacabum*) (Judd et al., 2002). Balloon vine is introduced to the eastern United States (USDA, NRCS, 2011).

#### (3) Discussion of Host Specificity Testing

*Parafreutreta regalis* reproduced readily on both the stipulate and exstipulate varieties of Cape-ivy occurring in California. *Parafreutreta regalis* did not cause gall formation or any signs of damage on any of the 103 species or varieties of plants, including 46 species in the tribe Senecioneae, that were tested (Balciunas et al., 2010; USDA, ARS, 2012). See appendix 1 for host specificity testing results. See appendix 3 for the permit applicant's proposed release strategy for *P. regalis*.

2. Impact of *P. regalis* on Cape-ivy
 Cape-ivy
 Two trials exposing Cape-ivy plants to two different densities of *P. regalis* were conducted in quarantine (Balciunas and Smith, 2006). After approximately two months, the growth of the galled vines to Cape-ivy vines that had not been exposed to *P. regalis* were compared. Under both the high density (10 pairs of flies/plant) and low density (2 pairs/

plant) treatments, the galled vines exhibited visible stunting, and the ungalled stems were longer, and had more nodes and larger leaves (Balciunas and Smith, 2006).

3. Uncertainties Once a biological control agent such as P. regalis is released into the Regarding the environment and becomes established, there is a slight possibility that it Environcould move from the target plants (Cape-ivy) to attack nontarget plants. mental Host shifts by introduced weed biological control agents to unrelated plants are rare (Pemberton, 2000). Native species that are closely related Release of P. to the target species are the most likely to be attacked (Louda et al., 2003). regalis If other plant species were to be attacked by P. regalis, the resulting effects could be environmental impacts that may not be easily reversed. Biological control agents such as P. regalis generally spread without intervention by man. In principle, therefore, release of this biological control agent at even one site must be considered equivalent to release over the entire area in which potential hosts occur, and in which the climate is suitable for reproduction and survival.

In addition, this agent may not be successful in reducing Cape-ivy populations in the continental United States. Worldwide, biological weed control programs have had an overall success rate of 33 percent; success rates have been considerably higher for programs in individual countries (Culliney, 2005). Actual impacts on Cape-ivy by *P. regalis* will not be known until after release occurs and post-release monitoring has been conducted. However, it is expected that *P. regalis* will reduce Cape-ivy populations by reducing growth rate and ability to climb, which are the main causes of the weed's impact on other vegetation.

- 4. Human Health It is a plant-feeding insect which poses no risk to humans or other animals.
- **5. Cumulative Impacts** "Cumulative impacts are defined as the impact on the environment which results from the incremental impact of the action when added to other past, present and reasonably foreseeable future actions regardless of what agencies or person undertakes such other actions" (40 CFR 1508.7).

Many state and national parks in California currently manage Cape-ivy. Golden Gate National Recreation Area recently spent \$600,000, over three years, trying to map and eradicate Cape-ivy. Currently Golden Gate and Point Reyes National Seashore are working to remove 188 acres of Capeivy patches in both parks. Removal of Cape-ivy using herbicides is underway in the Chorro Willows area of Morro Bay State Park. Cape-ivy is a problem along roadways, California's Department of Transportation (CalTrans) and CalTrans road crews devote time and herbicides for controlling this weed.

Release of *P. regalis* is not expected to have any negative cumulative

impacts in the continental United States because of its host specificity to Cape-ivy. Effective biological control of Cape-ivy will have beneficial effects for weed management programs, and may result in a long-term, non-damaging method to assist in the control of Cape-ivy.

# 6. Endangered Species Act Section 7 of the Endangered Species Act (ESA) and ESA's implementing regulations require Federal agencies to ensure that their actions are not likely to jeopardize the continued existence of federally listed threatened and endangered species or result in the destruction or adverse modification of critical habitat.

There are four federally-listed species in the plant family Asteraceae, tribe Senecioneae in the continental United States: *Blennosperma bakeri* (Sonoma sunshine), *Senecio franciscanus* (San Francisco Peaks ragwort) and its critical habitat, *Senecio layneae* (Layne's butterweed), and *Yermo xanthocephalus* (desert yellowhead) and its critical habitat. These listed plant species are the most closely related to Cape-ivy. Based on host specificity of *P. regalis* reported in host specificity testing and the literature, APHIS has determined that environmental release of *P. regalis* may affect, but is not likely to adversely affect the four listed plants and their designated critical habitats. APHIS submitted a biological assessment to the U.S. Fish and Wildlife Service (FWS) and requested concurrence with these determinations.

## V. Other Issues

Consistent with Executive Order (EO) 12898, "Federal Actions to Address Environmental Justice in Minority Populations and Low-income Populations," APHIS considered the potential for disproportionately high and adverse human health or environmental effects on any minority populations and low-income populations. There are no adverse environmental or human health effects from the field release of *P. regalis* and will not have disproportionate adverse effects to any minority or lowincome populations.

Consistent with EO 13045, "Protection of Children from Environmental Health Risks and Safety Risks," APHIS considered the potential for disproportionately high and adverse environmental health and safety risks to children. No circumstances that would trigger the need for special environmental reviews are involved in implementing the preferred alternative. Therefore, it is expected that no disproportionate effects on children are anticipated as a consequence of the field release of *P. regalis*.

EO 13175, "Consultation and Coordination with Indian Tribal Governments," was issued to ensure that there would be "meaningful consultation and collaboration with tribal officials in the development of Federal policies that have tribal implications...."

APHIS is consulting and collaborating with Indian tribal officials to ensure that they are well-informed and represented in policy and program decisions that may impact their agricultural interests in accordance with EO 13175.

## VI. Agencies, Organizations, and Individuals Consulted

The Technical Advisory Group for the Biological Control Agents of Weeds (TAG) recommended the release of *P. regalis* on April 25, 2013. TAG members that reviewed the release petition (USDA, ARS, 2012) included USDA representatives from APHIS, Agricultural Research Service, National Institute of Food and Agriculture, and the Forest Service; U.S. Department of Interior's Bureau of Reclamation and Bureau of Land Management; U.S. Army Corps of Engineers; Environmental Protection Agency; and representatives from Agriculture and Agri-Food Canada and Mexico.

This EA was prepared by APHIS and ARS. The addresses of participating APHIS units, cooperators, and consultants (as applicable) follow.

U.S. Department of Agriculture Animal and Plant Health Inspection Service Policy and Program Development Environmental and Risk Analysis Services 4700 River Road, Unit 149 Riverdale, MD 20737

U.S. Department of Agriculture Animal and Plant Health Inspection Service Plant Protection and Quarantine Regulations, Permits, and Manuals 4700 River Road, Unit 133 Riverdale, MD 20737

U.S. Department of Agriculture Agricultural Research Service Western Regional Research Center Exotic and Invasive Weeds Research Unit 800 Buchanan Street Albany, CA 94710

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**Appendix 1.** Results of *Parafreutreta regalis* "multi-choice/host added" host specificity experiments. Species in bold are probably sympatric with Cape-ivy in California or Oregon,  $^{SA}$  = plants native to S. Africa, \* = non-native species naturalized in USA. Number of genera/species from Bremer (1994) and USDA, NRCS (2011); families following Judd et al. (2002). (From USDA, ARS, 2012)

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Family Asteraceae (Order Asterales)				
Tribe: Senecioneae - 120 ge	enera worldwide, 2	1 in the US		
Subt	ribe Senecioninae -	68 genera worldwide, 7 in the US		
Cineraria sp. $(0 / 0)$				
<sup>SA</sup> C. "butterfly" ornamental cultivar hybrid	none	S. Africa, ornamental cultivar hybrid	Pretoria	0 / 8
<sup>SA</sup> C. deltoidea Sonder	none	S. Africa, coastal herb in S. Africa	Pretoria	0 / 5
<sup>SA</sup> C. saxifraga de Candolle	wild cineraria	S. Africa, ornamental in US	Pretoria	0 / 9
<i>Curio</i> sp. (0 / 0)				
C. rowleyanus (H. Jacobsen) P.V. Heath	string-of-pearls	S. Africa	Albany	0 / 5
<i>Delairea</i> sp. (2 / 2)				
*D. odorata Lemaire with stipules	Cape-ivy	widespread in coastal CA, SW OR	Both	131 / 131
*D. odorata Lemaire without stipules	Cape-ivy	widespread in coastal CA, SW OR	Both	6 / 6
<i>Erechtites</i> sp. $(8/4)$				
<i>*E. glomerata</i> (Desfontaines ex Poiret) de Candolle	cutleaf fireweed	Europe, West coast in US	Albany	0 / 5
Euryops sp. $(2/0)$				
<i>*E. chrysanthemoides</i> (de Candolle) B. Nordenstam	bull's eye	S. Africa, ornamental in US	Pretoria	0 / 9
*E. pectinatus (L.) Cassini	grey leafed euryops	S. Africa, ornamental in US	Both	0 / 10

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
*E. subcarnosus de Candolle	sweet resin bush	S. Africa, weed in Arizona	Albany	0 / 6
<i>Gynura</i> sp. (1 / 0)				
* <i>G. aurantiaca</i> (Blume) DC.	velvetplant, purple passion plant	Indonesia, ornamental, naturalized in Florida	Albany	0 / 5
<i>Mikaniopsis</i> sp. (0 / 0)				
<sup>SA</sup> M. cissampelina C. Jeffery	none	S. Africa, close relative of Cape ivy	Pretoria	0 / 5
<i>Packera</i> sp. (67 / 22)				
<i>P. bolanderi</i> (Gray) W.A. Weber & A. Löve	Bolander's ragwort	CA, OR, WA, west coast herb	Albany	0 / 10
<i>P. breweri</i> (Burtt-Davy) W.A. Weber & A. Löve	Brewer's ragwort	CA, woodland herb	Albany	0 / 5
<i>P. clevelandii</i> (Greene) W.A. Weber & A. Löve	Cleveland's ragwort	CA, North Coast Ranges, perennial	Albany	0 / 5
<i>P. ganderi</i> (Barkley & Beauchamp) W.A. Weber & A. Löve	Gander's ragwort	N. America, woodland herb, California listed – Rare	Albany	0 / 6
<i>P. macounii</i> (Greene) W.A. Weber & A. Löve	Siskiyou Mts. ragwort	N. America, widespread in US	Albany	0 / 5
Pericallis (1/1)				
<i>P. hybrid</i> (Regel) B. Nordenstam <i>Pseudogynoxys</i> sp. (1 / 1)	Florists cineraria	Africa, ornamental in the US	Albany	0 / 5
*P. chenopodioides Kunth	Mexican flamevine	S America, ornamental, formerly Senecio confusus	Albany	0 / 5

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Senecio sp. (86 / 34)				<b>1</b>
<sup>SA</sup> S. angulatus L. f.	garden senecio	S. Africa, ornamental	Pretoria	0 / 7
S. aronicoides DC.	rayless ragwort	CA, OR, biennial or perennial	Albany	0 / 5
<sup>SA</sup> S. articulatus (L.) C.H. "Bipontinus" Schultz	candle plant	S. Africa	Pretoria	0 / 5
S. blochmaniae E. Greene	dune ragwort	CA coastal sand dunes, subshrub	Albany	0 / 8
<sup>SA</sup> S. brachypodus de Candolle	none	S. Africa	Pretoria	0/6
<sup>SA</sup> S. deltoideus Lessing	canary creeper	S. Africa	Pretoria	0 / 5
S. flaccidus Lessing	threadleaf ragwort	widespread in southwest US, shrub	Both	0 / 10
<sup>SA</sup> S. gerrardii Harvey	none	S. Africa, widespread shrub	Pretoria	0 / 5
<sup>SA</sup> S. glastifolius L.f.	Waterdissel	S. Africa	Pretoria	0/6
<sup>SA</sup> S. helminthiodes (C.H. "Bipontinus" Schultz) Hilliard	none	S. Africa, widespread shrub	Pretoria	0 / 6
S. hydrophilus Nutt.	water ragwort	w. US, biennial or perennial	Albany	0 / 5
<i>S.integerrimus</i> Nutt. var. <i>exaltatus</i> (Nutt.) Cronquist	lambstongue ragwort	w. US, w Canada, biennial or perennial	Albany	0 / 5
*S. jacobaea L.	tansy ragwort	Eurasia, noxious weed in several US states	Albany	0 / 7
<sup>SA</sup> S. macroglossus de Candolle	flowering ivy	S. Africa, ornamental	Pretoria	0 / 5
S. mandraliscae (Tineo) H. Jacobsen	blue chalk sticks	Africa, formerly Kleinia mandraliscae	Albany	0 / 5

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
<sup>SA</sup> S. oxydontus de Candolle (form A)	none	S. Africa	Pretoria	0 / 9
<sup>SA</sup> S. oxydontus de Candolle (form B)	none	S. Africa	Pretoria	0 / 5
<sup>SA</sup> S. oxyriifolius de Candolle	false nasturtium	S. Africa	Pretoria	0 / 6
<sup>SA</sup> S. pleistocephalus S. Moore	none	S. Africa	Pretoria	0 / 5
<sup>SA</sup> S. tamoides de Candolle	canary creeper	S. Africa, widespread, ornamental	Pretoria	0 / 5
S. triangularis W. Hooker	arrowleaf ragwort	N. America, common in riparian areas common in riparian areas	Albany	0 / 5
*S. vulgaris L.	common grounsel	Europe & N. Africa, widespread US weed	Albany	0 / 7
Subt	ribe Blennospermat	tinae - 4 genera worldwide, 2 in the US		
<i>Blennosperma</i> sp. (4 / 4)				
<i>B. nanum</i> var. <i>nanum</i> (W. Hooker) Blake	common stickyseed	CA, uncommon herb	Albany	0/5
<i>Crocidium</i> sp. (1 / 1)				
C. multicaule Hook.	common spring- gold	CA, OR, WA, Br. Columbia	Albany	0 / 5
<u>Subt</u>	ribe Tussilagininae	- 48 genera worldwide, 12 in the US		
<i>Lepidospartum</i> sp. (3 / 2)				
L. latisquamum S. Watson	Nevada broomsage	CA, NV, UT, desert shrub	Albany	0 / 5
<i>Luina</i> sp. (2 / 1)				
L. hypoleuca Benthelot	littleleaf silverback	CA, OR WA, Br. Columbia, shrub	Albany	0 / 5

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Petasites sp. (7 / 2)				•
P. frigidus var. palmatus (L.) Fries	coltsfoot	w. US, Canada, perennial, common in riparian areas	Albany	0 / 5
Tribe: Anthemideae - 109	genera worldwide, 1	<u>3 in the US</u>		
<i>Achillea</i> sp. (20 / 9)	-			
A. millefolium L.	common yarrow	N. America, common herbaceous plant	Albany	0 / 6
<i>Artemisia</i> sp. (102 / 21)				
A. californica Lessing	coastal sagebrush	CA, common coastal shrub	Albany	0 / 6
Tribe: Arctoteae - 16 gener	ra worldwide, 6 in tl	ne US (All N. American species are intro	duced)	
Arctotheca sp. (1 / 1)				
*A. calendula (L.) Levyns	capeweed	S. Africa, noxious weed in CA	Pretoria	0 / 5
Tribe: Astereae - 170 gene	ra worldwide, 57 in	the US		
Baccharis sp. (25 / 12)				
B. pilularis de Candolle	coyotebrush	CA, OR, NM, common coastal shrub	Albany	0/6
<i>Erigeron</i> sp. (255 / 53)				
E. glaucus Ker-Gawler	seaside daisy	CA, OR, common coastal subshrub	Albany	0 / 5
<i>Grindelia</i> sp. (55 / 24)				
G. stricta de Candolle	Pacific gumweed	CA, OR, WA, common coastal subshrub	Albany	0 / 5
Symphyotrichum sp. (139 / 23)				
S. chilense (Nees) G.L. Nesom	Pacific aster	CA, OR, WA, Br. Columbia, formerly <i>Aster chilensis</i>	Albany	0/6

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Tribe: Calenduleae (Calend	lula officinalis & C	C. arvensis are the only representatives from	om this tribe i	n the US)
<i>Calendula</i> sp. $(2/2)$				
*C. officinalis L.	pot marigold	Eurasia, ornamental	Albany	0 / 6
Tribe: Cardueae - 83 generation	a worldwide, 11 in	the US		
Carthamus sp. (6 / 5)				
*C. tinctorius L.	safflower	Eurasia, safflower – commercial crop	Albany	0 / 6
Centaurea sp. (35/18)				
*C. melitensis L.	tocalote	Eurasia, weed in many states	Albany	0 / 5
<i>Cynara</i> sp. (2 / 2)				
*C. scolymus L.	globe artichoke	Europe, artichoke – commercial crop	Both	0 / 5
Tribe: Eupatorieae - 170 ge	enera worldwide, 20	6 in the US		
Ageratina sp. (20 / 4)				
*A. adenophora (Spreng.) R.M. King &	eupatory,	Mexico, ornamental & weed in US	Pretoria	0 / 6
H.E. Robins	crofton weed			
A. riparia (Regel) R.M. King & H.E. Robins	spreading snakeroot	Mexico, Caribbean, noxious weed in HI	Pretoria	0 / 5
Ageratum sp. $(4 / 1)$				
*A. houstonianum P. Miller	bluemink	C. America, se & ne US	Pretoria	0 / 5
<i>Campuloclinium</i> sp. $(0 / 0)$				
<sup>SA</sup> C. macrocephalum (Lessing) de Candolle	Pompom weed	S. America, invasive in S. Africa	Pretoria	0 / 5
Chromolaena sp. (9/9)	-			
<i>C. odorata</i> (L.) R.M. King & H.E. Robins	Siam weed	FL. TX. weed in HI	Pretoria	0/5
Mikania sp $(10/0)$		, ,		- · -
<sup>SA</sup> <i>M. capensis</i> de Candolle	none	S. America, S. African vine	Pretoria	0 / 4

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Tribe: Gnaphalieae - 180 ge	enera worldwide, 8	in the US		
Anaphalis sp. (1 / 1)				
<i>A. margaritacea</i> (L.) Bentham ex. C.B. Clarke	western pearly everlasting	N. America, widespread in the US	Albany	0 / 6
<i>Gamochaeta</i> sp. (6 / 5)				
G. purpurea (L.) Cabrera	spoonleaf purple everlasting	N. America, widespread weed	Albany	0 / 7
Tribe: Helenieae - 110 gene	ra worldwide, 71 ii	n the US		
Eriophyllum sp. (28 / 28)				
E. staechadifolium Lagasca y Segura	Seaside wooly sunflower, lizardtail	CA, OR, common coastal subshrub	Albany	0 / 6
<i>Madia</i> sp. (24 / 24)				
M. elegans D. Don ex Lindley	elegant madia	CA, OR, WA, NV, common grassland annual	Albany	0 / 5
Tagetes sp. $(8/2)$				
*T. erecta L.	Aztec marigold	C. America, ornamental - marigold	Albany	0 / 5
*T. minuta L.	wild marigold	S. America, noxious weed in CA	Pretoria	0 / 5
Tribe: Heliantheae - 189 ger	nera worldwide, 52	in the US		
<i>Cosmos</i> sp. (4 / 2)				
<sup>SA</sup> C. bipinnatus Cav.	cosmos, Mexican aster	<i>Bidens formosa</i> (Bonato) C.H. "Bipontinus" Schulz, non-native naturalized ornamental	Pretoria	0 / 2
Dahlia  sp.  (1 / 0)			D	0.1.6
*D. pinnata Cavanilles cv.	pinnate dahlia	C. America, ornamental	Pretoria	0/6

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Galinsoga sp. $(2/2)$				*
*G. parviflora Cavanilles	gallant-soldier	S. America, weed in US	Pretoria	0 / 5
Helianthus sp. (68 / 15)				
H. annuus L.	sunflower	N. America, sunflower – commercial crop	Pretoria	0 / 6
H. tuberosus L.	Jerusalem artichoke	N. America, Jerusalem artichoke	Pretoria	0 / 9
<i>Rudbeckia</i> sp. $(44 / 5)$				
R. hirta (garden cultivar)	blackeyed Susan	N. America, ornamental – coneflower	Pretoria	0 / 5
Zinnia sp. $(8/8)$				
*Z. violacea Cavanilles cv.	elegant zinnia	S. America, ornamental	Pretoria	0 / 5
Tribe: Inuleae - 38 genera	worldwide, 1 in the	US		
Dittrichia sp.				
*D. graveolens (L.) W. Greuter	stinkwort	Eurasia, minor weed in the US	Albany	0 / 5
Tribe: Lactuceae - 98 gene	ra worldwide, 32 in	the US		
Cichorium sp. (2 / 2)				
*C. intybus L.	chicory	Europe, chicory – minor crop and weed	Albany	0 / 8
<i>Lactuca</i> sp. (15 / 8)				
*L. sativa L.	garden lettuce	Europe, lettuce – commercial crop	Pretoria	0/6
<i>Picris</i> sp. (6 / 1)	-			
*P. echioides L.	bristly oxtongue	Europe, widespread weed in the US	Albany	0 / 5

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Tribe: Mutisieae - 76 gener	ra worldwide, 8 in t	he US		
Adenocaulon sp. (1 / 1)				
A. bicolor Hooker	American trailplant	N. America, common woodland herb	Albany	0 / 5
Tribe: Plucheeae - 28 gene	ra worldwide, 3 in	the US		
<i>Pluchea</i> sp. (14 / 3)				
P. odorata Cassini	salt marsh fleabane	N. America, widespread in US wetlands	Albany	0 / 6
Tribe: Vernonieae - 98 gen	era worldwide, 4 ir	<u>the US</u>		
Vernonia sp. (32 / 0)				
V. missurica Rafinesque	Missouri ironweed	central US, endangered in Ohio	Albany	0 / 6
Family Amaranthaceae (Order Caryophyllales)				
Beta vulgaris ssp. cicla (L.) Koch	chard	Europe, chard – commercial crop	Pretoria	0 / 5
Family Apiaceae (Order Apiales)				
*Hedera canariensis Willdenow	Algerian ivy	MAcronesia, ornamental vine	Albany	0 / 5
*Hedera helix L.	English ivy	Europe, ornamental vine, weedy	Albany	0 / 7
Family Aristolochiaceae (Order Piperales)		-		
Aristolochia californica Torrey	CA dutchman's pipe	N. America, including California	Albany	0 / 5
Family Brassiceae (Order Brassicales)				
*Brassica oleracea L.	cabbage	Europe, cabbage – commercial crop	Pretoria	0 / 5
*Lepidium latifolium L.	whitetop, pepperweed	Europe, noxious weed in several states	Albany	0 / 5
*Raphanus sativus L.	radish	Europe, radish – commercial crop	Pretoria	0 / 5

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Family Campanulaceae (Order Asterales)				
Campanula muralis L.	bellflower	Europe, ornamental in the US	Albany	0/6
*Lobelia erinus L.	edging lobelia	Europe, ornamental in the US	Albany	0/6
Family Cucurbitaceae (Order Cucurbitales)				
<i>Marah fabaceus</i> (Naudin) Naudin ex Greene	California manroot	N. America, common vine in California	Albany	0 / 6
Zehneria scabra (L. f.) Sonder subsp. scabra	none	S. Africa, vine	Pretoria	0 / 5
Family Ranunculaceae (Order Ranunculales)				
Clematis lingusticifolia Nuttall	Virgin's bower	N. America, common vine in western US	Albany	0 / 6
Family Rosaceae (Order Rosales)				
Fragaria chiloensis (L.) P. Miller	beach strawberry	N. America, common US west coast plant	Albany	0 / 8
Family Sapindaceae (Order Sapindales)				
Acer macrophyllum Pursh	big leaf maple	N. America, common US west coast tree	Albany	0 / 5
Acer negundo var. californicum (Torrey & A. Gray) Sargent	California box elder	N. America, common CA tree	Albany	0 / 5
Aesculus californica (Spach) Nuttall	buckeye	N. America, common California tree	Albany	0 / 5
Cardiospermum halicacabum L.	balloon vine	N. America ornamental vine	Albany	0 / 5
*Dodonaea viscosa (L.) Jacquin	Florida hopbush	N. America, ornamental, host of <i>Pa</i> . <i>mavoana</i>	Albany	0 / 5

Scientific name (No. of species, subspecies & varieties in the genus (in USA / in CA)	Common name(s)	Region of endemism, notes	Location of tests	Reps w/ adults / total reps
Family Vitaceae (Order Vitales)				
Vitis californica Bentham	California wild grape	N. America, widespread western US vine	Albany	0 / 5

#### Appendix 2. Host-specificity testing methods

#### **Field Surveys**

A field survey was conducted in South Africa during 1999 to search for prospective biological control agents on Cape-ivy in Western Cape, Eastern Cape and KwaZulu-Natal provinces (Grobbelaar, 1999). Six trips were conducted, and 60 sites were visited. In 2000, a survey was conducted to look for presence of *P. regalis* on closely related vines in the genera *Delairea* (1 species), *Senecio* (8 species), *Cineraria* (2 species), *Mikaniopsis* (1 species) and *Mikania* (1 species) (Grobbelaar, 2000). Ten sites were visited three times each during times of the year when the insect was most abundant. Plants at the sites were examined for presence of galls, and any galls were collected to rear out adult insects for identification.

#### Laboratory Host Specificity Experiments

The host range tests for *P. regalis* were performed from January 2001 through August 2011, under containment conditions in the greenhouse of the USDA-ARS weed biocontrol quarantine facility located at Albany, California, USA and in the laboratories of the Weeds Programme, Plant Protection Research Institute, in Pretoria, South Africa (Balciunas et al., 2010). During the tests conducted in the Albany quarantine, although supplemental heating and cooling were used, ambient temperatures were allowed to fluctuate between 11 and 36 °C. During winter, natural lighting was supplemented to maintain a 14-hour photoperiod by using four 200-watt incandescent bulbs, placed about 1.5 meters (m) above the plants. Test conditions in Pretoria were less controlled and supplemental lighting was occasionally used in winter.

The flies used in the tests were from laboratory cultures established with immatures collected near the town of Wilderness, in Western Cape Province, South Africa. The Cape-ivy plants used in Albany were grown from cuttings collected at Garrapata Creek, 15 kilometers south of Carmel, California. The Cape-ivy plants used in Pretoria were grown from cuttings collected at several locations in Kwazulu-Natal Province. The stipulate variety was primarily used in host range tests because it is the most common variety in both South Africa and California. Typically, the Cape-ivy control plants were grown in 20 centimeter (cm) (8 inch) diameter pots, had several stems, and the longest stem was about 0.4 m long. The form and size of the test plant species varied greatly, but they were in similar size pots, and all had at least one stem longer than 20 centimeters. Most of the test plants were grown from seeds, collected from the field as seedlings, or were propagated from cuttings by the applicant and colleagues. Some ornamental species were purchased from commercial sources. The scientific names for the North American species conform to those used by PLANTS Database (USDA, NRCS, 20011) while the names of South African plants conform to Arnold and de Wet (1993). The identity of representative specimens of each plant species tested in the Albany quarantine laboratory was confirmed by G. F. Hrusa and D. G. Kelch at California Department of Food and Agriculture, while those tested in Pretoria were identified by the staff of the Herbarium, South African National Biodiversity Institute, Pretoria. Insect voucher specimens have been deposited in the United States National Museum [USNM] at the Smithsonian in Washington D.C., in the California State Collection of Arthropods in Sacramento, and in the South African National Collection of Insects in Pretoria, South Africa.

A testing protocol was designed by the applicant ("multi-choice/host added") to maximize the data obtained during the short period that the adult flies were alive (about 2 weeks). Each test was begun with one plant each of four nontarget species, then a Cape-ivy plant was added on the fourth day to serve as a positive control. In Albany, experiments were done in a metal screen cage ( $122 \times 91\frac{1}{2} \times 91\frac{1}{2} \times 91\frac{1}{2} \times 201\frac{1}{2} \times 91\frac{1}{2} \times 201\frac{1}{2} \times 91\frac{1}{2} \times 91\frac{1}$ 

The host range tests conducted in Pretoria were nearly identical to those conducted in Albany. Three or four test plants of similar size were placed in a cage (0.56 m x 0.56 m x 0.6 m) for three days with four pairs of newly emerged flies. Insects were provided with a honey and yeast solution. On day four, the positive control, a Cape-ivy plant of similar size, was added. After three more days of exposure, the insects were removed, while the plants were left in the cage and monitored for damage and insect development. At both locations, if no galls were observed on the Cape-ivy control, then the test was considered 'unsuccessful', omitted from subsequent analyses, and repeated. Each plant species was tested at least five times.

In Albany, during 2003 and 2004, the applicant also conducted tests to determine if the gall fly had a preference for either of the two varieties of Cape-ivy (stipulate and exstipulate). These tests were similar to the host range tests described above and used the same cages. Four Cape-ivy plants (2 stipulate and 2 exstipulate), were placed in corners of the cage, and four pairs of P. regalis adults were released into the cage. In order to ensure more galls, another four pairs were added to the cage a week later. After 15-21 days, the dead and living flies were removed, and the Cape-ivy vines were monitored for gall development and adult emergence. After 85 days, all of the galls were cut off from the Cape-ivy vines, and dissected to count any larvae, pupae or dead adults. Each gall was classified into one of three classes: 1) 'Under-developed' galls – swellings at nodes induced by P. regalis oviposition (confirmed by dissection), but the larvae had died early, and these galls never reached their typical size; 2) 'Fully-developed without windows' – galls that reached a normal size, but 'windows' had never formed, and they contained dead larvae, but not pupae; 3) 'Fully-developed with windows' - galls from which adults sometimes emerged, and which, upon dissection, contained mixtures of dead and live larvae, and/or dead adults. The effect of variety of Cape-ivy on the numbers of each type of gall, or the number of adults and pupae produced in the galls, was analyzed using Student's *t*-test (Statistix, 2005). Prior to analyses, the count data was transformed by square root (x + 0.01) to improve normality (Snedecor and Cochran, 1967).

#### Appendix 3. Release Strategy for *P. regalis.*

The permit applicant identified two habitats in northern California to make initial releases: coastal regions of several Bay Area counties, and wooded regions in the East Bay hills (Alameda and Contra Costa counties). If the fly establishes at any of these sites, we will conduct studies to measure its impact, population growth and rate of dispersal.

*P. regalis* that would be released by the permittee come from one of the most southerly infestations of Cape-ivy in South Africa, near the town of Wilderness, at latitude of 34°S, which corresponds to the latitude of Los Angeles (34.5°N). Cape Agulhas, the southernmost point in South Africa (and the African continent) is only at latitude 34.8°S, so there is no place in Africa that corresponds in latitude to the Cape-ivy infestations around San Francisco (latitude 37.8°N) or those in southwestern Oregon (latitude 42.3°N). However, *P. regalis* was found at the high elevation sites in South Africa's Drakensberg Mountains, where winter snows are common, indicating that all infestations of this vine in California and Oregon have the potential of being attacked by this insect, if it is released.

The permittee plans to release 20 pairs of flies into field cages (approx. 6 x 8 x 5 feet) erected over a patch of Cape-ivy. The cages will be monitored weekly to detect signs of establishment (gall formation). After signs of emergence of the next generation of adult flies are apparent (after approximately 2 months) the cages will be removed, and the flies will be allowed to disperse naturally. If no establishment is evident after 3 months, supplemental releases of additional flies will be made into the cages.