A PETITION FOR THE INTRODUCTION AND FIELD RELEASE OF THE CHONDRILLA ROOT MOTH, BRADYRRHOA GILVEOLELLA (TREITSCHKE), FOR THE BIOLOGICAL CONTROL OF RUSH SKELETONWEED IN NORTH AMERICA

Submitted by
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ABSTRACT:

Rush skeletonweed, *Chondrilla juncea* L. (Asteraceae), is considered a noxious weed in many western states and is currently a target for biological control. *Bradyrrhoa gilveolella* (Treitschke) (Lepidoptera: Pyralidae) is a root-feeding moth being considered for use in the biological control of rush skeletonweed. This organism will serve to complement the existing biocontrol agents that have been established on rush skeletonweed in the Pacific Northwest but have provided only limited suppression of the weed. Although root feeders have suppressed populations of several noxious weeds in the western US, no root feeding organisms have been introduced into North America for this particular weed species. Host records (field) and laboratory studies indicate that the moth is restricted to the genus *Chondrilla*; and only *C. juncea* occurs in North America. Since the moth is genus specific there would be negligible risk to other related plant genera, including the closely related genera of *Taraxacum* and *Crepis*. We thereby recommend the release of this moth into North America.

I. INTRODUCTION

Nature of the Problem

1. History of Introduction and Spread.
Chondrilla juncea L. (rush skeletonweed) arrived in the eastern United States in the early nineteenth century as a weed seed in European grain (Reed 1979). In the western United States, rush skeletonweed was first reported near Spokane, Washington in 1938. Rush skeletonweed was reported in Idaho and Oregon in the 1960s and by 1995 infested over 6.2 million acres of rangeland in the Pacific Northwest and California (Sheley and Hudak 1995). It is estimated that the infestation of rush skeletonweed in the Pacific Northwest increases 41,000 ha per year (Spollen and Piper 1995). This weed was first reported in Montana in 1991 (Sheley and Hudak 1995).   Rush skeletonweed is currently increasing in the Pacific Northwest, as evidenced by the rise in number of infested counties from close to zero in 1938 to over 50 in 1999 (New Invaders Database 1999). Viewing a plot of these figures, rush skeletonweed appears to be on an exponential increase in the Northwest and may indicate the potential for rapid spread in western North America.

2. Present Distribution in North America.
The present distribution of rush skeletonweed in the United States includes the following states: California, Delaware, Georgia, Idaho, Indiana, Maryland, Michigan, Montana, New Jersey, New York, Oregon, Pennsylvania, Virginia, Washington, Washington D.C. and West Virginia (USDA-NRCS 1999, BONAP/MIP 1999). Rush skeletonweed is also present in Canada; being found in Ontario as well as in British Columbia (British Columbia 1999).

3. Sectors Affected and Magnitude of the Problem.
Rush skeletonweed presently infests over 6.2 million acres of rangeland in the Pacific Northwest and California and is currently invading British Columbia and Montana. Infested land types include roadsides, railways, rangelands, pastures, grain fields, coastal sand dunes, and shaley hillsides in mountainous regions (Reed 1979; Sheley and Hudak 1995). In Australia , competition from rush skeletonweed reduced wheat yields by as much as 80%, resulting in estimated losses of more than $25 million (18.6 million due to lost of production and 1.4 million for direct control) (Cullen 1986). Although no similar economic studies on estimated losses in North America are known to us, we predict

a similar loss in infested grain fields and additional losses due to dramatically reduced rangeland forage production.

4. Consensus that the Weed is a Suitable Target for Control.
Rush skeletonweed has not been placed on the Federal Noxious Weed List but has been listed as noxious by the following states/provinces: Arizona, California, Colorado, Idaho, Montana, Oregon, South Dakota, Washington, and British Columbia (New Invaders Database 1999, USDA-NRCS 1999). However, it should be noted that many state noxious weed lists are not very inclusive and tend not to include species which are not currently present in the state but which would pose a threat if introduced. Thus, rush skeletonweed is a larger threat than the current state weed lists reflect. Because of its importance as a weed, a biological control program was undertaken against rush skeletonweed in the 1970s that resulted in the successful introduction of three biological control agents. By the 1980s, these agents produced satisfactory control of rush skeletonweed in California and some areas of Washington. However, these agents do not provide effective control over most of the North American range of rush skeletonweed, particularly the cooler, interior lands of Oregon, Washington, Idaho, and Montana. As a result, a new program is underway to find, import, and establish a complex of new biological control agents capable of controlling rush skeletonweed in North America.

Proposed Action

We are requesting approval for the release of the of the root boring moth *Bradyrrhoa gilveolella* (Treitschke) to augment the biological control of rush skeletonweed in western North America.

II. TARGET WEED INFORMATION

Taxonomy of the Target Weed

1. Classification.

Phyllum: Magnoliophyta
    Class: Magnoliopsida
        Order: Asterales
            Family: Asteraceae
                Subfamily: Cichorioideae
                    Tribe: Lactuceae
                        Subtribe: Crepidinae
                            Genus: *Chondrilla*
                                Species: *juncea* L.

Common name: rush skeletonweed (approved common name - North American), also hogbite, gum succory, devil's grass and naked weed.

2. Identifier.
Rush skeletonweed was identified by Linnaeus and bears the scientific name *Chondrilla juncea* L. There are no synonyms for this species.

3. Problems in identification or taxonomy.
There are no problems in identification or taxonomy for rush skeletonweed, although biotypes or forms of the plant are known to exist. In Southern Europe, 300 forms are said to have been recognized (Cullen 1991), of which at least three have been introduced into western North America (see Distribution of the Target Weed. 5. Information on Genetic Variability).

4. Origin and location of herbarium specimens and the date of depository.
The New Invaders Database (http://invader.dbs.umt.edu/queryplant1.asp) lists 206 distribution records (herbarium and others) for rush skeletonweed in the Pacific Northwest. No species priority site has been identified which maintains germplasm of *Chondrilla juncea* L. (USDA-ARS 1999). The USDA Forest Service Rocky Mountain Research Station is currently maintaining voucher herbarium specimens of *Chondrilla juncea* L. accessions used in their host specificity studies.

Description of the Target Weed

Rush skeletonweed (Fig. 1) is a herbaceous biennial or perennial that can grow up to 1.3 meters (four feet) tall with spreading side branches (Whitson et al. 1991, Holm et al. 1997). Taproots can penetrate to a depth of 2.4 meters in the soil and new plants arise from lateral root buds in the upper 0.6 meters of soil. The taproot remains more or less the same thickness for its entire length and divides only occasionally in the upper layers of soil. Fine branch-roots occur at intervals and lateral roots may be produced in the surface layers of the soil. Rosettes resemble dandelions and can grow to diameters of 37 cm or more. Rosettes vary from dark green to purplish in color, especially in the fall. During late spring, a spindly stem elongates from the center of the rosette. At this time, the basal leaves have deep, irregular teeth that generally point back to the stem base. The stem leaves are narrow, generally linear, and relatively inconspicuous giving the plant a skeleton-like appearance. A key species characteristic is the presence of downwardly bent, coarse reddish hairs on the lower four to six inches of the stem. All plant parts exude a thick, white latex sap when wounded. The bright yellow flowerheads develop along the stem and branch tips either singly or in clusters of two to five flowerheads. Flowerheads are ¾ inch in diameter and composed of seven to fifteen individual florets with a very fine, soft pappus of bright white bristles. First year plants usually produce 250 to 350 seeds, but older plants can produce over 20,000 seeds. The light brown or black ribbed, pappus-bearing seeds are about 1/8 inch long.

Distribution of the Target Weed

1. Native Range.
The genus *Chondrilla* is considered to have its center of origin in Central Asia (Caresche and Wapshere 1974). Rush skeletonweed is native to Eurasia and ranges from the Iberian Peninsula through southern Europe, Asia Minor and the Mediterranean/Caspian Sea region to the Altai Mountains eastward to Mongolia and to Algeria and Tunisia in North Africa (Reed 1979, USDA-ARS 1999).

FIGURE 22-2 *Chondrilla juncea* L.: 1. basal leaves showing root system and lower part of stem with bristles; 2. tip portion of plant, showing narrow leaves and inflorescences; 3. seedling; 4. various leaf forms; 5. fully mature flower head; 6. seed with stalked parachute-like pappus.

Taken from Holm et al. 1997.
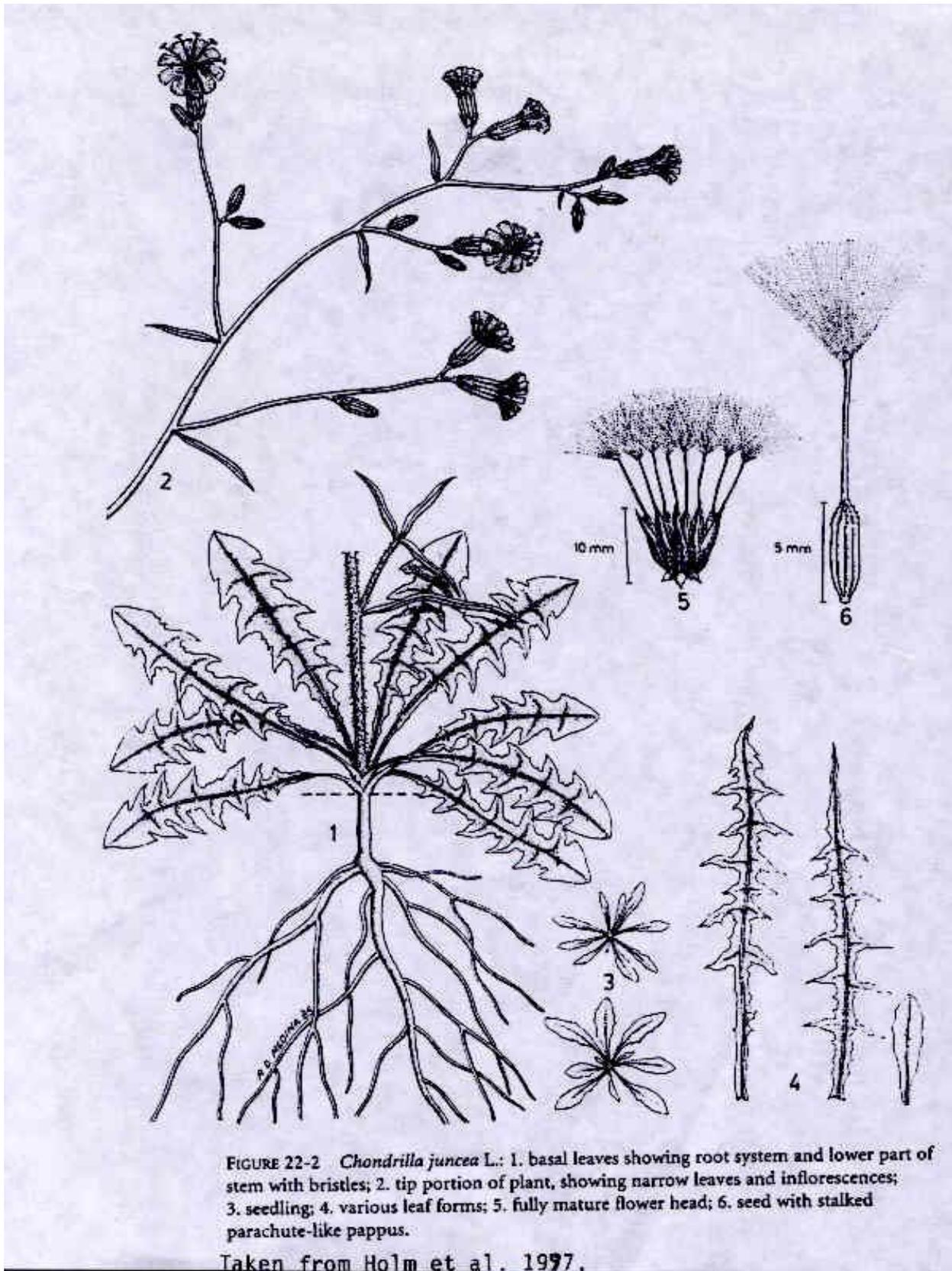
Figure 1. *Chondrilla juncea* L.

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2. Worldwide Areas of Introduction, Pattern of Movement, and Limit (see Fig. 2).
Rush skeletonweed has successfully invaded Australia, Argentina, Canada, New Zealand, and the
United States (Holm et al. 1997, USDA-ARS 1999).

FIGURE 22-1 The distribution of *Chondrilla juncea* L. across the world where it has been
reported as a weed.
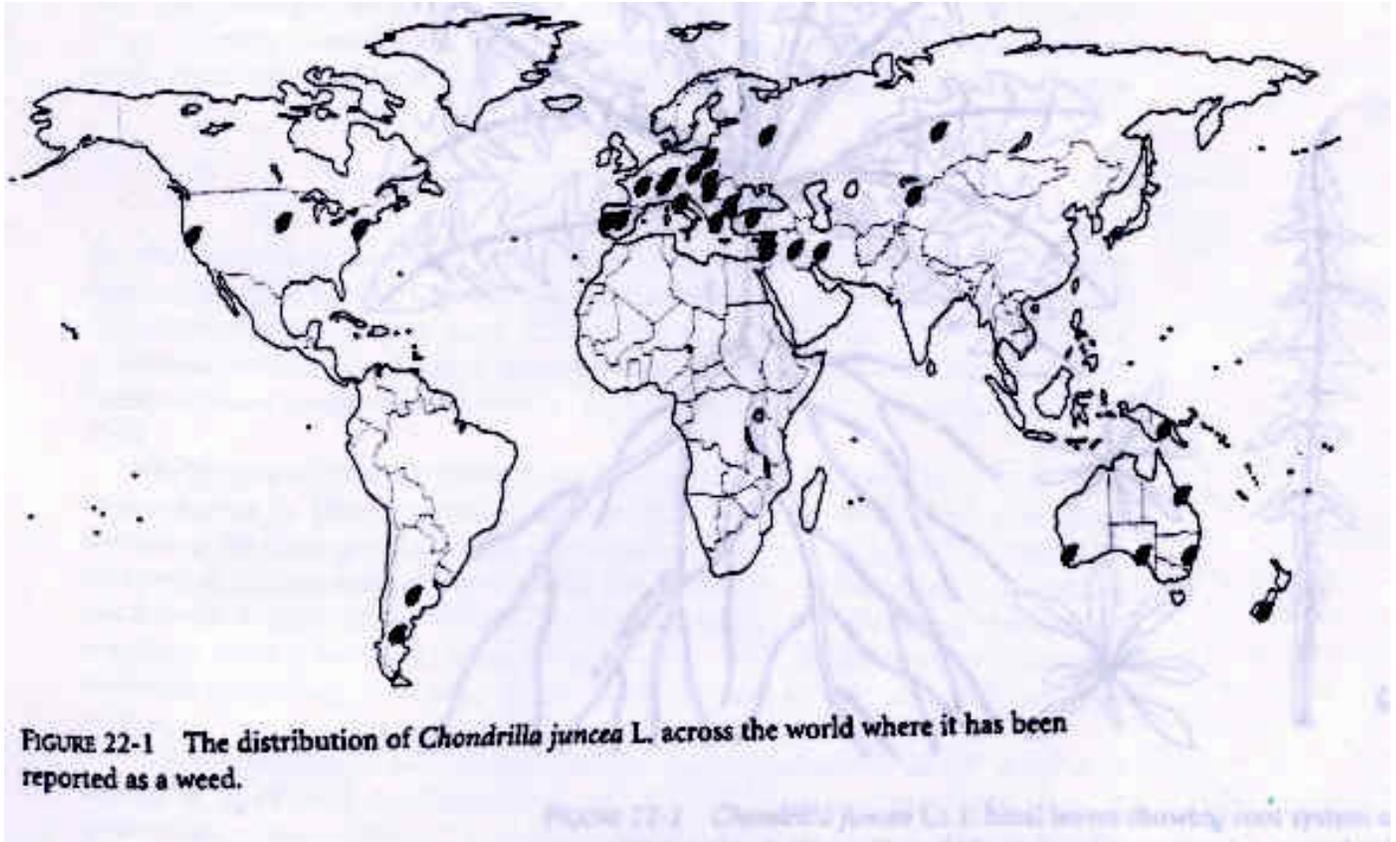
Figure 2. World wide distribution of *Chondrilla juncea* (Holm et al. 1997).

3. Present distribution in North America (see Fig. 3).
The present distribution of rush skeletonweed in the United States includes the following states:
California, Delaware, Georgia, Idaho, Indiana, Maryland, Michigan, Montana, New Jersey, New York,
Oregon, Pennsylvania, Virginia, Washington, Washington D.C. and West Virginia (BONAP/MIP 1999,
USDA-NRCS 1999). In Canada, rush skeletonweed is present in Ontario and in British Columbia,
where it is designated a provincial noxious weed (BONAP/MIP 1999, British Columbia 1999).

4. Area of Potential Spread in North America.
Rush skeletonweed has a wide tolerance for varying climatic conditions, as evidenced by its widespread
distribution. Winter extremes appear to range from those areas with little or no frost to others where
minimums are in the -20 $^{\circ}$C range. Summer temperatures reaching 59 $^{\circ}$F (15 $^{\circ}$C) appear to be
necessary for flower and seed production (McVean 1966). Precipitation in infested areas varies between
23 and 152 cm (McVean 1966, Lee 1986). Thus, there appears to be no biological reason why rush
skeletonweed could not survive in Canada and other northern states. In Eurasia, the species ranges from
35 to 55 $^{\circ}$ N latitude and elevations of sea level to 1800 m (McVean 1966). Rush skeletonweed is

currently increasing in the Pacific Northwest, as evidenced by the rise in number of infested counties from close to zero in 1938 to over 50 in 1999 (New Invaders Database 1999). Viewing a plot of these figures, rush skeletonweed appears to be on an exponential increase in the Northwest and may indicate the potential for rapid spread in western North America.
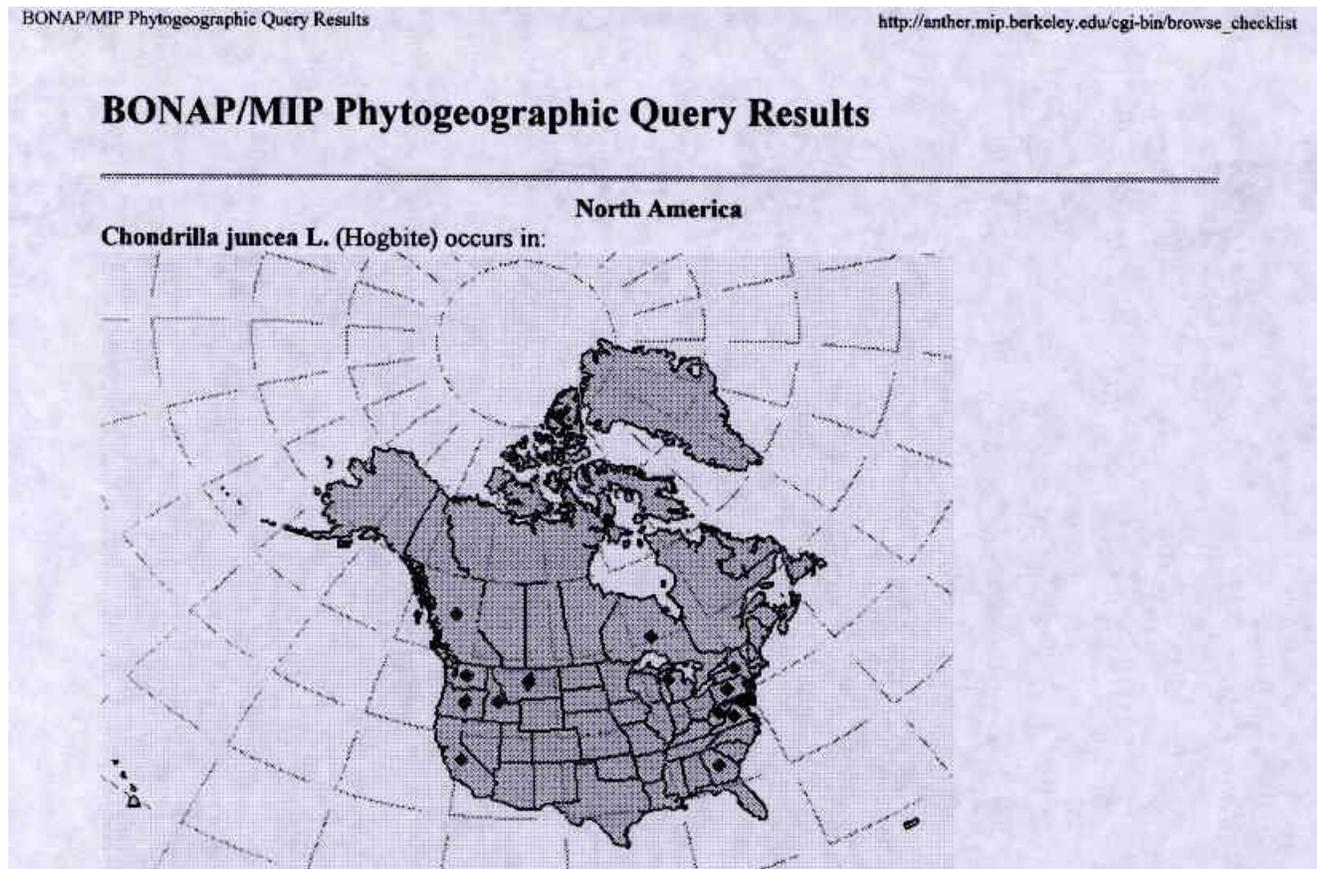
Figure 3. North American distribution of *Chondrilla juncea* (BONAP/MIP 1999, USDA-NRCS 1999).

5. Information on Genetic Variability.

Rush skeletonweed shows a preference for well-drained, light sandy or gravel soil types. As these soil types tend to be well separated through the original range of Eurasia, individual populations have evolved more or less in isolation to form genetically distinct biotypes (Caresche and Wapshere 1974). In Southern Europe, 300 forms are said to have been recognized (Cullen 1991). Evolution of biotypes is facilitated by the plant's reproductive character. Rush skeletonweed is a triploid and has been described as an obligate apomicitic, meaning it produces seeds by division of the germ cells without the necessity for fertilization, thus each individual plant is identical to its parent (Rosenberg 1912). This enables a highly competitive genotype to maintain itself without change, and eventually allowing the genotype to dominate an area. New biotypes may develop from autosegregation or by mutation (Burdon et al. 1980).

Most researchers report at least three or four "varieties" or "biotypes" in the Western United States with distinct differences in plant height, branching patterns, phenology of flowering, and susceptibility to the *Puccinia* rust (Littlefield 1980, Lee 1997). Isoenzyme analysis of North American and European genotypes by Hasan et al. (1995) suggest three distinct biotypes of rush skeletonweed in North America. Genotype I ("Washington - late flowering") is located in northern Idaho and Washington, but is also present in California and Oregon, Genotype II ("Washington - early flowering") is located in northern Idaho and Washington, and Genotype III ("Banks") is in central Idaho. Comparisons with European rush skeletonweed suggest that genotypes identical to the "Banks" and "Washington – late flowering" are also located at several sites in Yugoslavia.

6. Habitats/Ecosystems Where Weed is Found in North America.
Rush skeletonweed is adapted to a variety of the following conditions: 1) semi-arid climates with cool winters and warm summers where severe droughts do not occur; a precipitation pattern with intermittent spring and fall rains; 2) well-drained, rather acidic gravely, sandy soils or dry, shallow rocky soils; xeric to mesic sites; 3) warm aspects well exposed to sunlight such as open south-facing slopes; 4) grasslands in disclimax; 5) open forest types; and 6) areas where disturbances have exposed and/or degraded the site (McVean 1966, Wapshere et al. 1974, Wapshere et al. 1976). Native stands of vegetation in good condition are seldom invaded by rush skeletonweed, although grasses are poor competitors once the weed is established. In Montana, big sagebrush/needle and threadgrass, bluebunch wheatgrass/ Sandberg's bluegrass, and bitterbrush/bluebunch wheatgrass are some of the habitat types thought to be susceptible to invasion (Sheley and Hudak 1995). In British Columbia, ponderosa pine, bunchgrass, interior Douglas fir, and interior cedar-hemlock habitats are predicted to be at high risk for infestation. In North America, rush skeletonweed infests roadsides, railways, rangelands, pastures, grain fields, coastal sand dunes, and shaley hillsides in mountainous regions (Whitson et al. 1991).

Taxonomically Related Plants and Their Distribution

There are about 25 species of *Chondrilla* worldwide. They are primarily located in central and southern Europe, North Africa, the Middle East, Iran, Afghanistan, Pakistan, Central Asia, and China (Bremer 1994). There are no *Chondrilla* species native to North America, and only *C*. juncea has been introduced.

Life History of the Target Weed

Rush skeletonweed is a herbaceous, long-lived perennial. Seeds germinate in the fall and produce seedlings and small rosettes that persist over winter. In the western states, and even under the less severe Australian climate, top growth of mature plants dies to the ground as soon as they are affected by frost, although fall rosettes may develop given adequate moisture (McVean 1966, Littlefield 1980). Growth resumes in the spring with one to several new rosettes forming at the crown of the parent plant with one or more main stems arising from each rosette. By early summer the basal leaves gradually wither and later in the summer season, the narrower stem leaves are also shed, leaving the plant to photosynthesize by stems alone. Flower production occurs from maturity in early summer until terminated by frost (Littlefield 1980). Seeds start to mature three days after flowering and are readily dispersed by wind, water, and animals, including humans within 10 to 20 days (McVean 1966, Cuthbertson 1970). Mature plants can produce over 20,000 seeds annually. Seeds require no dormancy period for germination and germinate at temperatures between $7\,^{\circ}C$ (45° F) and $40\,^{\circ}$ C ($104\,^{\circ}$ F) with

optimal germination at 25 °C (77 °F) (McVean 1966). Seeds may remain viable for up to four years (Cuthbertson 1970). Seedlings require continuous moisture for three to six weeks for successful establishment (Cullen and Grove 1977). During drought, most seedlings die after emerging (McVean 1966). In addition to reproducing through seeds, rush skeletonweed can vegetatively produce shoots from rhizomes and by regeneration following rootstock fragmentation. Shoots can arise from pieces as short as 1 cm and from as deep as 1 m below soil surface and regeneration following injury can occur at any time of year (McVean 1966, Holm et al. 1997). If plants are damaged by drought, new rosettes form whenever effective rain is available and almost immediately send up flowering stems.

Impacts of the Target Weed

1. Beneficial Uses.
Rush skeletonweed is a somewhat drought-tolerant pasture plant that is palatable and nutritious for sheep in the rosette and early flowering stage, and is a grazed component of low quality pastures in many parts of southeastern Australia. When rain is adequate, rush skeletonweed can be a major source of pollen for honeybees (Sheley and Hudak, 1995). We are not aware of any medicinal or herbal uses for rush skeletonweed. Prior to World War II, rush skeletonweed was investigated in Russia as a potential source of rubber, but it was determined that not enough latex was produced by the plant for it to be considered as an economical alternative (Iljin 1930).

2. Social and Recreation Use.
Rush skeletonweed does not have any known social or recreational use. Rush skeletonweed monocultures are considered by most people to be aesthetically unpleasant in comparison to healthy ecosystems.

3. Impact on Threatened and Endangered Species.
We know of no studies that have determined the effects of rush skeletonweed invasion on threatened and endangered species. However, it can be assumed that rush skeletonweed would affect threatened, endangered, and sensitive species because of its ability to form dense monocultures that compete for soil moisture and nutrients; thereby displacing indigenous species.

4. Economic Losses.
Rush skeletonweed presently infests over 6.2 million acres of rangeland in the Pacific Northwest and California (Sheley and Hudak 1995) and is currently invading British Columbia and Montana. Competition from rush skeletonweed reduced wheat yields by as much as 80% in Australia, resulting in estimated losses of more than $25 million (Cullen 1986). Although we know of no economic studies on estimated losses in North America, we predict a similar loss in infested grain fields and additional losses due to dramatically reduced rangeland forage production. The ecological losses in plant and animal diversity are also enormous but cannot be economically calculated. Additional expenditures result from control costs.

5. Health Dangers.
No human or animal health dangers have been associated with rush skeletonweed.

6. Regulatory Aspects.
Rush skeletonweed has not been placed on the Federal Noxious Weed List but has been listed as noxious in Arizona, California, Colorado, Idaho, Montana, Oregon, South Dakota, Washington, and British Columbia (New Invaders 1999, USDA-NRCS 1999). Regulations vary by state/province but generally involve actions such as restricted importation and/or quarantine and prevention, containment, eradication or other control measures.

7. Effects on native plant and animal populations.
We know of no studies that have determined the effects of rush skeletonweed invasion on native plant and animal populations. However, it can be assumed that rush skeletonweed does affect native plants and animals because of its ability to form dense monocultures that compete for soil moisture and nutrients, thereby displacing indigenous species.

8. Impact of current weed control on non-target plants
Conventional measures such as herbicides and cultural controls are non-selective and themselves affect non-target plants. For example, picloram, when used to control rush skeletonweed, can severely injure or kill many desirable non-target plants, including trees. Because picloram is relatively persistent, it may injure plants for several growing seasons. Picloram can also leach through sandy soils or be lost in surface water runoff and contaminate streams and groundwater.

9. Effect on ecosystem functions and ecological relationships
As rush skeletonweed monocultures invade species-rich range and mountain habitats, ecosystem functions and ecological relationships are affected. Rush skeletonweed generally negatively impacts plant and animal diversity.   No studies that address the effects of rush skeletonweed on nutrient cycling and disturbance regimes, such as fire and flood frequencies, are known to us. However, given rush skeletonweed's ability to dominate a community, it is reasonable to assume that this species does affect nutrient cycling and disturbance regimes. Any change in diversity and/or nutrient cycling and disturbance regimes resulting from the introduction of an exotic plant may be viewed as detrimental to a native ecosystem.

Alternative Management Options

1. Historical Options.

 Rush skeletonweed is difficult to control with herbicides. Historically, picloram (Tordon 22K®) has been applied at 2 quarts per acre to rosettes. 2,4-D amine at a rate of 2 quarts per acre may also provide some control (Sheley and Hudak 1995).   In Australia, a single application of clopyralid (Stinger®, 1.5 pints per acre) reduced rush skeletonweed shoots approximately 60% three years after application. Mixing clopyralid (Stinger®, 1.5 pints per acre) with dicamba (Banvel DMA®, 2 quarts per acre) or picloram (Tordon 22K®, 1 quart per acre) plus 2,4-D (2 quarts per acre) gave the best long term control. Successful control depends on specific conditions of the site and usually requires reapplication on an annual basis. However, while the expense of repeated application may be justifiable on high return cereal crops, it is unlikely to be cost effective in range situations.

Cultural control practices have also been used to control rush skeletonweed in selected infestations. Hand pulling or grubbing provides effective control of small infestations, but must be repeated several

times during the growing season over several year period. Mowing and annual cultivation does not control rush skeletonweed. Mowing does not affect carbohydrate reserves, although in dry years it may limit seed production (Sheley and Hudak 1995). Low level cultivation may increase infestations by creating and spreading root fragments, whereas intensive cultivation every six to eight weeks may eliminate the weed (Holms et al. 1997). The effect of rush skeletonweed on wheat and pasture yields may be minimized under moist conditions by increasing competition by the addition of high rates of nitrogen fertilizer.   Also planting competitive legumes, such as alfalfa, increases soil fertility and effectively reduced populations of rush skeletonweed in crop-pasture rotations. However, the high level of pasture management needed for effective control is difficult to achieve. Competitive plantings of legumes combined with the presence of the rust has proven effective in augmenting the control of rush skeletonweed in Australia (Groves and Williams 1975). Proper grazing by sheep may reduce or prevent production of rush skeletonweed (Cuthbertson 1966). Continuous, rather than rotational grazing, produces the lowest weed densities. Moderate grazing is as effective as heavy grazing; as heavy grazing decreases the competitive ability of desired species.

Three biological control agents have been released on rush skeletonweed in North America. A rust, *Puccinia chondrillina*, causes pustules that erupt through the leaf and stem surfaces which desiccate the leaves and reduce the plant's ability to photosynthesize (Hasan 1972). Severe rust infections can control rush skeletonweed under certain conditions, whereas light infections may reduce seed production and viability. A gall mite, *Aceria chondrillae*, induces the vegetative and floral buds to form leafy galls (Caresche and Wapshere 1974). Severe galling may cause stunting of the plant and greatly reduces seed production. A gall midge, *Cystiphora schmidti*, feeds on the rosettes, stem leaves, and stems, deforming plants and reducing seed production (Littlefield 1980). Gall midges have less impact than either the rust or mites and are subjected to high levels of parasitism. Although the effectiveness of individual biocontrol agents may vary as to the local, in California the rust appears to be more effective (Supkoff et al. 1988), whereas in eastern Washington the mite appears to be more important (Spollen and Piper 1995).

2. Current Options.
Early detection and herbicide treatment is critical in preventing the establishment of the weed at new locations. Where infestations are well established, an integrated approach utilizing several control techniques may be necessary to achieve the long-term control of rush skeletonweed. The use of competitive plantings, grazing by sheep or other animals, and biological control appears to have potential for managing rush skeletonweed infestations.

3. Potential Options
While the three biological control agents have been established over most of the range of rush skeletonweed in California, Idaho, Oregon, and Washington, they have not provided effective control in many areas. A new program is underway to determine if new natural enemies exist, if cooperators can be found, and if sufficient funding will be available to implement the biological studies necessary to find, import, and establish a complex of new biological control agents capable of controlling rush skeletonweed.

III. BIOLOGICAL CONTROL AGENT INFORMATION

Taxonomy of Bradyrrhoa:

1. Common Names: Chondrilla root moth (unofficial)

2. Classification:

Phylum: Arthropoda
    Class: Insecta
        Order: Lepidoptera
           Suborder: Ditrysia
              Superfamily: Pyraloidea
                Family: Pyralidae
                  Subfamily: Phycitinae
                    Genus: *Bradyrrhoa*
                      Species: *gilveolella* (Treitschke)

3. Synonymy: Not available.

4. List of closely related taxa in North America
The genus *Bradyrrhoa* is comprised of more than 20 species, all of which are of Mediterranean or western Asian origin. No species are known to occur in North America.

5. Identification of Bradyrrhoa

    <u>Who identified agent, names and locations</u> - *Bradyrrhoa gilveolella* was originally described by Treitschke in 1832.

    <u>Description of life stages</u> - Adult and larval descriptions are provided in Kozulina and Rudakova (1932). Adults are 11-13 mm in length, 25-28 mm in wingspan, and are creamy buff color with the anterior wing having three brown bands across it. Larvae are stippled and with few bristles. Initially the body is pink with a brown head, but later changes to a cream color (Caresche and Wapshere 1975). Mandibles are tridented with an anterior cutting lobe. Crochets, except for the anal pair, are arranged in a biordinal, uniseries circle. Mature larvae are 20-26 mm in length, and width of the head capsule is 1.6-2.6 mm. Pupae are light brown and finely foveolate but with a smooth appearance. Length ranges from 11 to 14 mm and width is approximately 3 mm. Eggs are prolate spheroids, 0.65 – 0.80 mm in length, and 0.45 mm in diameter. When laid, eggs are a creamy white color but darken with age. Larvae may be seen through the reticulated chorion prior to hatching.

Problems in identification - No problems are anticipated with the identification of *Bradyrrhoa gilveolella*. Only one other root-boring moth (*Ena* (= *Oporopsamma*) *wertheimsteini* (Rebel) (Lepidoptera: Tortricidae) is associated with rush skeletonweed. *Ena wertheimsteini* feeds in the upper root crown and is readily distinguished from the phyticid. In addition, *E. wertheimsteini* is not known to occur in eastern Greece (Hasan and Wasphere 1977), although it could potentially be found in northern or western Greece.

Voucher specimens – Voucher specimens are located at the British Museum, Natural History Museum (US), and at the Quarantine Lab, Montana State University.

Who identified the Agent – P. Wholly, British Museum.

Reason for choosing agent

Bradyrrhoa gilveolella was selected as a potential agent for several reasons: 1) larvae attack the roots of rush skeletonweed causing the death of smaller plants, interrupting nutrient flow of larger plants, and making the root of the plant more susceptible to invasion by pathogenic fungi; 2) no other root feeding organism has been introduced into North America for the control of rush skeletonweed; 3) damage by the moth will augment control achieved by other biological control agents; 4) the moth may have two generations per year which overlap and may cause damage through the growing season, increasing the plant's susceptibility to dry summer conditions; and 5) the moth appears to be host specific to rush skeletonweed.

Geographical Range:

1. Native Range
Records prior to 1968 indicate that *B. gilveolella* is present in Southern Russia from Kazakhstan to the Ukraine, extending westward into Turkey, Romania, Bulgaria, Yugoslavia, Macedonia, and Sicily (Caresche and Wapshere 1975). In Caresche and Wapshere's (1975) study, *B. gilveolella* was found throughout eastern mainland Greece and in northern and central Iran, Azerbaidjan and Karadj.

2. Other Areas of Introduction
 Bradyrrhoa gilveolella has been released but not successfully established in Australia and Argentina (Julian and Griffiths1998).

 3. Expected range in North America
 Establishment of the moth is expected in many areas of northwestern North America having rush skeletonweed, especially in areas of lighter soil types. Based upon observations made by Caresche and Wapshere (1975) of the persistence of southern Greek populations of *B. gilveolella*, moth populations may not be able to persist in parts of California or in eastern North America, where climatic conditions may not be as conducive.

4. Geographical range of closely related taxa in North America
Species of *Bradyrrhoa* are not known to occur in North America.

Life History:

1. Biology

Larval development and pupation take place entirely beneath the soil surface within a feeding tube that is attached to the root of *Chondrilla*. The case is made of loosely spun silk to start with but later it is covered with latex, root fragments, frass, and soil particles (Caresche and Wapshere 1975). The developmental stages of *B. gilveolella* in Caresche and Wapshere's (1975) study were as follows: egg 6-10 days; larva 45-60 days; and pupa 7-10 days. Adults emerge via their tubes. Two to eight days after emergence the females will lay eggs. Kozulina and Rudakova (1932) (in Caresche and Wapshere 1975), suggest that the female, under field conditions, will lay her eggs on the soil at the base of *Chondrilla* plants. When kept in laboratory cages, females will lay eggs on the walls, as well as on the stems of the *Chondrilla* plants. Newly emerged larva will descend to the soil by a silk thread, and crawl across the soil surface until they encounter a plant. The larva will briefly feed on the stem before attaching to the root 5-10 mm below the soil surface. A similar biology was observed in the laboratory by Littlefield et al. (unpublished data).

The Russian populations have overlapping generations. One generation occurs from May/June to September and the other from late July, early September until May/June. The larva of the winter generation remains dormant from November to March (Kozulina and Rudakova 1932, in Caresche and Wapshere 1975). Caresche and Wapshere (1975) found that in northern Greece the life cycle is similar to the Russian populations. The adults of the overwintering generation emerge in May and June and the summer generation emerges in August to October. The two generations are not as distinct in Greece due to the shorter winter. The southern Greece population cycle is similar to the one in the north with the overwintering population emerging slightly earlier, in late April to early May (Caresche and Wapshere 1975).

2. Known Mortality Factors

Caresche and Wapshere (1975) found *B. gilveolella* to be parasitized by several species of Hymenoptera and also by a Tachinid fly. *Copidosoma* sp. (Hymenoptera: Encyritidae) was the most common parasite, infesting up to 30% of the moths. *Syzeuctus* sp. (Hymenoptera: Ichnemonidae) was found parasitizing up to 10% of the population, with *Bracon* sp. (Hymenoptera: Braconidae) being the least important. The Tachinid fly, *Germaria graeca*, was widespread throughout the population but usually at a small rate (5 %) of parasitism. Two fungi, *Cordyceps* sp. and *Beauveria bassiana* were found to infect the larvae (Caresche and Wapshere 1975), with *Cordyceps* being of little importance. *Beauveria bassiana* is the most important biological factor affecting larval populations of *B. gilveolella*. The combination of Hymenoptera and fungi can cause a 50-70% reduction in larval populations with the Tachinid fly attacking 20-40% of the remaining pupae.

In quarantine studies in Bozeman, several species of parasitoids emerged and were tentatively identified (based on the Australian work in Greece) as: *Syzeuctis* sp. (Ichnemonidae), *Bracon* sp. (Braconidae), *Copidosoma* sp. (Encyritidae) and *Germaria graeca* (Tachnidae). A *Peltochalcidia* sp. also emerged but is considered to be a hyperparasite of the tachnid. Parasitoid emergence was estimated to have occurred from 7 to 12% of the larvae/pupae collected in 1997. Similar levels of parasitism were also observed in 1988 and 1999. No pathogens were found in association with larval or adult *Bradyrrhoa*.

3. Known Host Range (Specificity):

Field Observations – *Bradyrrhoa gilveolella* has only been found, under field conditions, infesting other *Chondrilla* species (L'Homme 1935, Caresche and Wapshere 1975). It is found on *C. juncea*, *C. juncea* form *intybacea* (= *C. latifolia*), *C. brevirostris*, *C. ambigua*, *C. kossinskyi* (= *C. pauciflora*), *C. kusnezovii*, and *C. mujunkumensis* in southern Russia (Sakharov 1930; Kozulina and Rudakova 1932; Dirsch 1933). Caresche and Wapshere (1975) found *B. gilveolella* on *C. juncea*, *C. juncea* form *acantholepis* (= *C. acantholepis*), and *C. ramosissima* in Greece.

The only other species of *Bradyrrhoa* for which the host plant is known, is *B. lyratella* Chretien found on *Andryala lyrata* (Asteraceae: tribe Lactuceae) (L'Homme 1935, in Caresche and Wapshere 1975).

Laboratory Observations (Host Specificity Testing) -

Populations of the Agent Studied **-** The earliest studies on the biology of *B. gilveolella* were conducted in southern Russia. Later studies on the biology and host specificity were conducted in Greece and the United States.

Sites of Study - Biological studies and host specificity testing by Caresche and Wapshere (1975) were conducted in southern and northern Greece. Host specificity testing of the Greek population of *B. gilveolella* was conducted by Littlefield et al. at the Insect Quarantine Laboratory located at Montana State University, Bozeman.

Experimental Methodology, Analysis and Host Range Testing

Previous Host Specificity Testing   (Caresche and Wapshere 1975)

Purpose **-** Although field collections and host records indicate that *B. gilveolella* feeds exclusively on *Chondrilla* spp., studies were conducted to demonstrate the host specificity and safety of this moth for importation into Australia for biological control.

Test Plant List - *Bradyrrhoa gilveolella* was tested against 77 plant species in 21 plant families (Table 1). Sixteen species of Asteraceae, of which seven were in the Lactuceae, were tested. Most of the plants are of economic importance to Australia and were suggested by plant quarantine authorities (Caresche and Wapshere 1975).

Agent Source **-** Larvae for testing were collected from several sites located in Greece. Larvae were collected primarily from *C. acantholepis*.

Target Source - Rush skeletonweed plants collected from Greece were used as controls. For determining the adaptation of *B. gilveolella* to various *C. juncea* forms or biotypes, five forms were used: three Australian forms, the French "Aniane", and the Greek "*acantholepis*" (= *C. acantholepis*), which was used as a control.

Replicates - See methods section below.

Methods - Two methods were used to determine the host specificity of *B. gilveolella*. The first was an "in vivo" test in which medium to large larvae (8-20 mm in length) were extracted from field collected plants and then placed on roots of growing plants or near root portions of plants. Three to six larvae were used per plant and four plants of each species were tested. Rush skeletonweed was used as a control. Plants were maintained at (18° C night, 26° C day). After 15 days, the top portion of the root crown was inspected for feeding, and after one month the entire root system was inspected for larval feeding. All tested species of Asteraceae (except *Taraxacum officinale* and *Sonchus oleraceus*) were treated in this manner.

A second method, "in vitro", was used for the remainder of the test plants, plus *Cichorium endivia* and *Lactuca sativa*. Six to ten larvae were placed in a 25 cm diameter glass petri dish lined with filter paper on the bottom. One dish, for a no-choice test, had three 15 cm pieces of the test roots placed in the dish. The other dish, for the choice test, had two root sections of the test plant located between pieces of the rush skeletonweed control. Dishes were covered with a piece of glass and were maintained at 20-25° C, 12:12 h (D:L) lighting. After 24 hours the plate was slid slightly off allowing ventilation of the plate but not allowing the roots to dry out. Each test was run for seven days. After two days roots were inspected and at the end of the experiment, plants were checked for signs of feeding or the attachment of the larvae to the root.

Results - No feeding was observed on any of the test plants except for rush skeletonweed in the "in vivo" tests. Larvae rapidly attacked the roots of rush skeletonweed, spinning their webbed feeding tubes. In most cases typical feeding tubes were completely constructed by the end of the experiment. For two species, *Ficus carica* and *Pinus radiata*, larvae used bits of bark material to add to their web but did not feed upon the plants. For *Saccharum officinarum*, larvae entered the pith through cut openings in the crown. Larvae utilized the pith in their webbing but did not feed on healthy root epidermis. Larvae on all other plant species were dead after 30 days.

In the "in vitro", no-choice tests, *Taraxacum officinale* roots were fed upon as with the rush skeletonweed controls. Larval feeding on *T. officinale* proceeded at a much slower rate compared to *C. juncea*. Larval mortality on all other plant species started after 7 days. In choice tests, all larvae had attached themselves to rush skeletonweed after one week. Feeding or attachment of larvae was not observed for other plant species.

To determine if possible feeding on *T. officinale* occurred under field conditions in Greece, 250 plants in the vicinity of infested rush skeletonweed plants were dug and inspected. None of the rootstock showed indications of feeding by *B. gilveolella* larvae.

Tests on the three Australian and two European forms (biotypes) of *C. juncea*, indicated no differences in feeding acceptance by *B. gilveolella*.

Table 1. Host test plant list – Testing by Caresche and Wapshere 1975.

_____

Asteraceae		
Cichorioideae		
Lactuceae		
Crepidinae	*Chondrilla juncea*	Rush skeletonweed
	Taraxacum officinale	Dandelion
Lactuicinae	*Lactuca sativa*	Lettuce
Scorzonerinae	*Scorzonera hispanica*	
Sonchinae	*Sonchus arvensis*	Corn sowthistle
	S. oleraceus	
unassigned	*Cichorium endivia*	Endive
Cardueae	*Carthamus tinctorius*	Safflower
	Cynara scolymus	Artichoke
Asteroideae		
Anthemideae	*Chrysanthemum indicum*	
	C. leucanthemum	
Helenieae	*Tagetes* sp.	
Heliantheae	*Helianthus annuus*	Sunflower
	H. tuberosus	Jerusalem artichoke
	Dahlia sp.	
	Zinnia sp.	
Calenduleae	*Calendula* sp.	
Brassicaceae	*Brassica oleracea*	Cabbage
	B. rapa	Turnip
Chenopodiaceae	*Beta vulgaris*	Beet
Convolvulaceae	*Ipomoea batatas*	Sweet potato
Cucurbitaceae	*Cucurbita maxima*	Pumpkin
	Cucmis sativus	Cucumber
	C. melo	Rock melon
	Citrullus vulgaris	Water melon
Fabaceae	*Pisum sativum*	Garden pea
	Phaseolus vulgaris	French bean
	Vicia faba	Broad bean
	Glycine hispida	Soy bean
	Medicago sativa	Lucerne
	Trifolium subterraneum	Subterranean clover
	T. repens	White clover
	Acacia dealbata	Wattles
	A. floribunda	
	Medicago tribuloides	Barrel medic
	M. littoralis	Strand medic

Table 1. Continued.

Juglandaceae	*Juglans regia*	Walnut
Liliaceae	*Asparagus offinalis*	Asparagus
	Allium cepa	Onion
Linaceae	*Linum usitatissimum*	Linseed, flax
Malvaceae	*Gossypium* spp.	Cotton
Moraceae	*Ficus carica*	Fig
Myrtaceae	*Eucalyptus globulus*	Gum
	E. camaldulensis	Gum
Oleaceae	*Olea europaea*	Olive
Pinaceae	*Pinus radiata*	Monterey pine
Poaceae	*Triticum* spp.	Wheat
	Hordeum vulgare	Barley
	Avena sativa	Oats
	Secale cereale	Rye
	Oryza sativa	Rice
	Zea mays	Maize
	Sorghum vulgare	Sorghum
	Saccharum officinarum	Sugar cane
	Lolium perenne	Perennial ryegrass
	Phalaris tuberosa	Phalaris
Rosaceae	*Malus sylvestris*	Apple
	Pyrus communis	Pear
	Prunus domestica	Plum
	P. persica	Peach, nectarine
	P. armeniaca	Apricot
	P. cerasus	Cherry
	P. amygdalus	Almond
	Cydonia vulgaris	Quince
	Fragaria vesca	Strawberry
	Rosa spp.	Garden rose
Rutaceae	*Citrus sinensis*	Orange
	C. limonia	Lemon
	C. paradisi	Grapefruit
Solanaceae	*Solanum tuberosum*	Potato
	Lycopersicum esculentum	Tomato
	Nicotana tabacum	Tobacco
	Capsicum annuum	Capsicum
Umbeliferae	*Daucus carota*	Carrot
	Pastinaca sativa	Parsnip
	Apium graveolens	Celery
Urticaceae	*Humulus lupulus*	Hop
Vitaceae	*Vitis vinifera*	Grape

Host Specificity Testing   (Littlefield, Birdsall, Markin and Helsley 1997-1999)

Purpose - Although extensive host specificity tests were conducted by Australian researchers (Caresche and Wapshere 1975), the potential of *Bradyrrhoa gilveolella* to feed on closely related North American plant species (or genera) still needed to be resolved, thus host specificity tests were conducted at the Quarantine Laboratory, Montana State University from 1997 to 1999.

Test Plant List - Although Caresche and Wapshere (1975) conducted previous host specificity tests in Europe, we thought it necessary to test 27 plant species, along with several additional subspecies and varieties, to determine the potential for *Braddyrhoa gilveolella* to feed on North America species and genera closely related to rush skeletonweed. In developing our host specificity test list, we followed the phylogenetic approach suggested by Wapshere (1974), where closely related species are theorized to be at greater risk of attack than distantly related species. In selecting plants, we concentrated on closely related species with morphological characteristics similar to rush skeletonweed, i.e. perennial, tap-rooted species. We also identified biochemically related plant species and addressed concerns for threatened, endangered, and sensitive plant species in North America. Appendix I provides a detailed explanation for our selection of test plants, following the format suggested in the Reviewer's Manual for the Technical Advisory Group for Biological Control Agents of Weeds. We believe that by testing these 27 species, in combination with the previous host specificity information collected by Caresche and Wapshere (1975), we are able to infer if *Braddyrhoa gilveolella* is likely to attack any non-tested North American plant species.

Agent Source **-** 1997- Two collections of *Bradyrrhoa gilveolella* were made from three locations in northern Greece by J. Kashefi, USDA-ARS, EBCL. The first collection was made between June 6-8 at Aspravalta Road and Nea Apolinia Road (between Thessaloniki and Kavala), and Prespansko Jerero (Lake Prespa) NW of Florina. These collections were received at the MSU Quarantine on 23 June. A second collection was made on 6 & 7 July at Prespa Lake and received in quarantine 9 July.

1998- Two collections of *Bradyrrhoa gilveolella* were made from Prespansko Jerero (Lake Prespa) in northwest Greece by J. Kashefi, USDA-ARS, EBCL. The first collection was made between 25-27 May and was received at the MSU Quarantine on 11 June. A second collection was made on 23-25 July, and was received in quarantine 30 July.

1999 – A collection of *Bradyrrhoa gilveolella* was again made from Prespansko Jerero (Lake Prespa) in northwest Greece by J. Kashefi, USDA-ARS, EBCL on 19 & 29 May and was received at the MSU Quarantine on 8 July.

Target Source **-** Rush skeletonweed plants were collected from Pullman, Whitman County, Washington ("Washington late-flowering" – hereby designated WLF) (control plants); Banks, Boise County, Idaho ("Banks"); Liberty Lake, Spokane County, Washington ("Washington early-flowering"); and Placer and Nevada Counties, California ("Washington late-flowering"). These collections are representative of the major "biotypes" of rush skeletonweed in the western United States (see Distribution of the Target Weed. 5. Information on Genetic Variability).

Replicates - Between five and sixteen replicates per plant species were used. When possible, test plants were inoculated in groups of five individuals (per species) and an additional five plants were inoculated at a later date depending upon the availability of larvae and/or plants. A rush skeletonweed control (WLF) (five plants) was used as a control. A new set of control plants was used with each new cohort of larvae used.

Methods:

Plants - Plants, either as seeds or roots, were grown in Sunshine Mix [®] within 15 or 17 cm diameter pots. Plants were placed in a greenhouse, at 15-25° C with a natural photophase or on a day-long cycle, until roots were of suitable size to support larval development. To reduce larval mortality due to excessive soil moisture the sunshine mix around the base of the plant was removed (4- 5 cm diameter and depth) and replaced with sand just prior to inoculation with larvae.

Rearing - Roots with attached feeding tubes of *Bradyrrhoa gilveolella* were collected in Greece. These roots primarily contained larvae of various instars or pupae, although many were devoid of insects. Infested roots were placed in moist vermiculite in 10x20 propagation trays. Trays were covered with domed lids, vented at either end and then placed in isolation cages to contain any unwanted organisms. The trays were inspected for adult emergence, and the vermiculite and roots were periodically moistened.

1997- Approximately 250 feeding tubes were present in shipment 1 and 350 tubes in shipment 2. Adult emergence from these tubes was approximately 20% for both shipments (51 adults from shipment 1 and 73 adults from shipment 2). The adult sex ratio was nearly even (1.2 Γ: 1.0 E).

1998 - Approximately 338 infested roots were present in shipment 1, and 178 roots in shipment 2. The adult emergence from these roots was poor, with only 27 adults eclosing from shipment 1 and eight adults from shipment 2.

1999 – Approximately 265 roots were received. Many of the moths apparently had already emerged and only 15 adults were reared from the remaining roots.

Oviposition – Upon emergence, adult moths were introduced into a 60 x 60 x 60 cm cage containing 5-6 rush skeletonweed plants for mating and oviposition for the start of a rearing colony. Adults were provided with water and a nutrient solution (honey water and/or Gatorade [®]). Cages were placed in a containment greenhouse at 15-25° C and with a natural photophase. Most of the moths were removed from the cages after two or three days and were placed in 15 cm x 9 cm diameter acetate tubes. Tubes were lined with a quilted paper towel and the ends were sealed with vented caps. Vials containing water and water/honey solution or Gatorade [®] were provided. Approximately 6-8 adults were introduced per tube. The sex of the adult was difficult to determine without excessive handling, so adults were randomly selected.   Tubes were placed in a quarantine greenhouse at 15-25° C, with a natural photophase and inspected daily for eggs. Water and nutrient solutions were refreshed as needed.

The use of acetate tubes provided the best means to obtain eggs. Eggs that were laid on plants in rearing cages were difficult to locate due to their small size and coloration (or lack of initial coloration). In the tubes nearly all the eggs were laid in dimples of the quilted paper towels; often multiple eggs were laid per dimple. Eggs hatched from 7-10 days. Approximately 75% of the eggs laid were viable, of which 3% died prior to hatching.

Host Testing - Ten neonate larvae obtained from lab rearings were transferred to each test plant with a fine brush. Larvae were placed near the base of the plant, either among the rosette leaves, the leaf-stem axils or within the upper root crown. Five replications per test species were made and each test was repeated twice given adequate numbers of plants and insects. A rush skeletonweed (WLF) control was used for each group of plants tested. Plants were arranged in a randomized block design within a greenhouse maintained at 15-25° C and with a natural photophase. After approximately 60 days, or at the time of plant death, roots were carefully removed from the pot and separated from the soil. Feeding tubes and larvae/pupae were counted, and prior feeding or root damage was recorded for each plant.

Results:

Results 1997 - Fourteen different plant species, as well as three varieties of lettuce and five collections of rush skeletonweed were tested in 1997. No noticeable feeding or feeding tubes were found on any of the test plants except for rush skeletonweed (Table 2). Larvae seem to accept all biotypes of rush skeletonweed provided. Approximately 64% of the plants (both control and test rush skeletonweed) were infested with an average of 3.3 larvae or pupae per plant. All larvae were mid to late instar and several pupae were located within the feeding tubes at the time of harvest. Feeding by larvae extended from the base of the rosette to a maximum depth of 10 cm. The larvae fed on the outer cortex of the root causing a groove in the plant tissue. This feeding groove was as much as 2 mm in width and extended 2-3 mm into the root. Depending upon the size of the root, feeding was superficial to extensive, with up to half of the root diameter consumed. In several cases the root had begun to rot at the site of feeding and a *Fusarium* sp. (fungi: Hyphomycetes) was isolated from the root.

Results 1998 - Sixteen different plant species were tested in 1998 (Table 2). One hundred and thirty five replications of test plants were harvested. No feeding or feeding tubes were found on any of the test plants except for rush skeletonweed and *Lygodesmia juncea.* Only one small feeding tube (no feeding or larvae) was found on *Lygodesmia juncea*. All 15 replications of rush skeletonweed were infested with larvae, with an average of 6.8 larvae/plant. Larval development at the time of harvest varied from mid instar larvae to nearly mature individuals.

Results 1999 – Two *Crepis* species, *C. acuminata* and *C. atribarba* were tested, along with a rush skeletonweed (WLF) control. The two *Crepis* species were field collected and were beginning to senesce at the time of inoculation. Plants that appeared to be in poor condition were harvested early before the roots began to rot. No feeding or feeding tubes were observed on any of the *Crepis* roots. In contrast, 100% of the rush skeletonweed plants were attacked, with an average of 1.8 larvae per root.

Table 2. Host specificity testing of *Bradyrrhoa gilveolella* - Larval Development: 1997-1999 studies.

Family: Tribe Species	Subtribe	Year Tested	# Reps Tested	# Plants w/ Feeding	#Tubes (# Larvae or Pupae)
Asteraceae: Lactuceae					
Chondrilla juncea (WLF) (control)	Crepidinae	1999	6	6	14 (11,0)
Chondrilla juncea (WLF) (control)	Crepidinae	1998	15	15	112 (102,0)
Chondrilla juncea (WLF) (control)	Crepidinae	1997	16	8	30 (20,12)
Chondrilla juncea (Banks, ID)	Crepidinae	1997	5	3	11 (11,1)
Chondrilla juncea (Nevada Co., CA)	Crepidinae	1997	5	4	12 (6,3)
Chondrilla juncea (Placer, Co., CA)	Crepidinae	1997	5	4	11 (11,1)
Chondrilla juncea (Liberty Lake, WA)	Crepidinae	1997	5	4	11 (11,0)
Crepis acuminata	Crepidinae	1999	7	0	0
Crepis atribarba	Crepidinae	1999	6	0	0
Crepis elegans	Crepidinae	1998	5	0	0
Crepis runcinata	Crepidinae	1997	10	0	0
Taraxacum eriophorum	Crepidinae	1998	10	0	0
Taraxacum laevigatum	Crepidinae	1998	5	0	0
Taraxacum officinale	Crepidinae	1997/98	10	0	0
Taraxacum officinale ssp.*ceratophorum*	Crepidinae	1998	5	0	0
Agoseris aurantiaca	Microseridinae	1998	10	0	0
Catananche caerulea	Catananchinae	1997	10	0	0
Cichorium intybus	Not assigned	1997	10	0	0
Hieracium albertinum	Hieraciinae	1998	10	0	0
Lactuca sativa (Romaine)	Lactucinae	1997	10	0	0
Lactuca sativa (Grand Rapids)	Lactucinae	1997	10	0	0
Lactuca sativa (Iceberg)	Lactucinae	1997	10	0	0
Lactuca serriola	Lactucinae	1997	10	0	0
Lactuca tartarica var. *pulchella*	Lactucinae	1997/98	10	0	0
Lactuca virosa	Lactucinae	1998	10	0	0
Lygodesmia juncea	Stephanomeriinae	1998	15	0	1 (0,0)
Prenanthes sagittata	Lactucinae	1997/98	10	0	0
Sonchus oleraceus	Sonchinae	1998	10	0	0
Sonchus uliginosus	Sonchinae	1997	10	0	0

Table 2. Continued.

Family: Tribe / Species	Subtribe	Year Tested	# Reps Tested	# Plants w/ Feeding	#Tubes (# Larvae or Pupae)
Asteraceae: Lactuceae					
Stephanomeria tenuifolia	Stephanomeriinae	1997	10	0	0
Asteraceae: Arctoteae					
Gazania splendens x *rigens* (Harlequin hybrid)		1997	10	0	0
Asteraceae: Cardueae					
Cirsium undulatum		1997/98	10	0	0
Asteraceae: Mutisieae					
Gerbera jamesonii		1998	5	0	0
Asteraceae: Vernonieae					
Stokesia laevis		1998	10	0	0
Fabaceae:					
Vigna radiata		1997	10	0	0

Discussion

Host specialization –

Bradyrrhoa gilveolella has only been found, under field conditions, infesting rush skeletonweed and other *Chondrilla* species (Sakharov 1930; Kozulina and Rudakova 1932; Dirsch 1933, L'Homme 1935, Caresche and Wapshere 1975). This root-feeding moth has been found infesting *C. juncea*, *C. juncea* form *intybacea* (= *C. latifolia*), *C. brevirostris*, *C. ambigua*, *C. kossinskyi* (= *C. pauciflora*), *C. kusnezovii*, and *C. mujunkumensis* in southern Russia (Sakharov 1930; Kozulina and Rudakova 1932; Dirsch 1933). Caresche and Wapshere (1975) found *B. gilveolella* on *C. juncea*, *C. juncea* form *acantholepis* (= *C. acantholepis*), and *C. ramosissima* in Greece. In addition, laboratory tests conducted by Caresche and Wapshere (1975) and Littlefield et al. in 1997-1999 support field observations that *B. gilveolella* is host specific to the genus *Chondrilla*. In North America no other species of *Chondrilla* occur, thus none would be at risk. Both laboratory studies indicate that larvae are able to feed and develop on various biotypes of *C. juncea*, consequently there would no need to search for additional strains of the moth for adaptation to North American forms of the weed.

Potential Impact on Native Species –

No *Chondrilla* species other than rush skeletonweed are present in North America (Kartesz and Kartesz, 1980, USDA-NRCS 1999). The most closely related species in North America belong to two genera in the same subtribe as rush skeletonweed (Crepidinae): *Taraxacum* and *Crepis*. Of the two, *Taraxacum* is more closely related to *Chondrilla* compared to *Crepis* (Bremer 1994).

There are seven to nine *Taraxacum* species (depending on the species concept used) in North America. Most are western in distribution, although a few are arctic or cosmopolitan. We tested four *Taraxacum*

taxa (Table 2), including the introduced dandelions, *T. officinale* and *T. laevigatum*, which are cosmopolitan in distribution and polymorphic in character. We also tested *T. officinale* ssp. *ceratophorum* (syn. *T. ceratophorum*) and *T. eriophorum*, which are native. None of these species were fed upon by first instar larvae. However, Caresche and Wapshere (1975) did observe feeding on *Taraxacum officinale* under laboratory conditions in Europe. This feeding could be attributed to their use of mid to late instar larvae, which may be able to utilize a broader range of hosts. It has been observed for other moth species that later instar larvae are able to utilize a greater variety of hosts compared to early instar larvae (Reavy 1993). Caresche and Wapshere did not state whether these late instar larvae were able to complete their development on *T. officinale* but did say that they were unable to locate larvae infesting *T. officinale* at their field sites. They confirmed the contention of the Russian workers Sakharov (1930) and Kozulina and Rudakova (1932) that *B. gilveolella* is specific to *Chondrilla* species under natural conditions. In Southern Russia, Pravdin (1957: cited by Caresche and Wapshere 1975) also noted that *Taraxacum* did not serve as a field host for *B. gilveolella*. During the study of various Cichoriaceae, including two Old World *Taraxacum* species, *T. koksaghyz* and *T. multiscaposum*, *B. gilveolella* was never found on *Taraxacum* species nor on any other Cichoriaceae except *Chondrilla* species. Based on these observations and studies and our tests on four *Taraxacum* taxa, we are confident that *Taraxacum* species would not be physiologically suitable as hosts for *B. gilveolella* and would not be selected for oviposition by female *B. gilveolella* due to differences in plant morphology compared to bolted rush skeletonweed.

The other genus of Crepidinae, *Crepis*, has 23 species in North America, primarily in the western United States and Canada. Of the 23 species, 17 are biennial or perennial and 6 are annuals. We tested four perennial, taprooted species of *Crepis* (Table 2) with a wide distribution in western North America as we believe only the biennial and perennial species would be able to support overwintering populations of *B. gilveolella*. None of the species tested were fed upon by first instar larvae. Since no feeding, even rudimentary, was observed on any of the tested *Crepis* species, we believe there is little risk of attack on non-tested *Crepis* species.

In our tests, 25 Asteraceae species were tested, including 11 native species. At least one species was tested from each of the six subtribes native to the continental Untied States which are in the same tribe as rush skeletonweed, as well as one species from a subtribe with introduced species and one species from an unassigned genus. At least one species was also tested from each of the three tribes native to the continental United States in the same subfamily as rush skeletonweed, along with one species from a tribe with introduced species. In addition, Caresche and Wapshere (1975) tested at least one species from three of the eight tribes native to the continental United States but in different subfamilies than rush skeletonweed. No larval feeding or development was observed on any species other than rush skeletonweed. Although one small feeding tube was observed on *Lygodesmia juncea*, no feeding was associated with it. This probably occurred because, although *Lygodesmia juncea* is very similar morphologically to rush skeletonweed, it differs in its biochemistry. We believe that the combined tests on these 37 Asteraceae species, along with the additional tests on non-Asteraceae species, present solid evidence that *B. gilveolella* is unlikely to present a risk to any species, native or introduced, other than *Chondrilla*.

Potential Impact on Threatened, Endangered, or Sensitive Species -

The Asteraceae is one of the largest plant families in North America and, accordingly, has a high number of threatened, endangered, and sensitive species (TES). With threatened and endangered species in more than 20 genera and sensitive species in over 70 additional genera, the number of potential test species is prohibitively large. Instead, we followed Wapshere's phylogenetic theory that risk would be highest to those species most closely related to rush skeletonweed and limited our testing to those species within the same tribe as rush skeletonweed (Lactuceae). In North America, there are seven subtribes of Lactuceae with native species; five of these subtribes contain TES species. Because obtaining seeds and/or plant material of TES species can be difficult and may further decimate populations, we decided not to test the species of concern. After reviewing the 11 listed species, we selected a substitute species for each from the same genera, except *Microseris* where we tested an *Agoseris* species (Table 3). The criteria used for selection of substitutes was that the species be perennial so that larvae might complete their development by overwintering, have a taproot large enough for feeding tube formation, and have a similarly shaped rosette that might attract an ovipositioning female.

The two species considered to be most at risk and that received our highest attention because they fell in the same subtribe as rush skeletonweed are *Taraxacum californicum* and *T. carneocoloratum*. *T. californicum* is found in California, possibly within the range of rush skeletonweed, and is listed as endangered (USFWS 1999). *Taraxacum carneocoloratum*, historically found in Alaska and the Yukon, outside the current range of rush skeletonweed, was formerly listed as a Category 2 species (USFWS 1993). (Note - In 1996, the U. S. Fish and Wildlife Service discontinued the use of this category, but remains concerned about and acknowledges the need for further study of these sensitive species (USFWS 1996)). Based on the observations and studies of *Taraxacum* species described above and our tests on four *Taraxacum* taxa, we believe no *Taraxacum* species, including any TES taxa, would be suitable hosts for *B. gilveolella*.

Four subtribes in the same tribe as rush skeletonweed contain TES species in the continental United States (USFWS 1993, USFWS 1996). The Lactucinae has two species of *Prenanthes* formerly listed as Category 2, *P. barbata* and *P. bootii*. The Microseridinae has two species of *Microseris* formerly listed as Catergory 2, *M. decipens* and *M. howellii*. The Stephanomeriinae has two species formerly listed as Category 2, *Lygodesmia dolorensis* and *Stephanomeria blairii,* and one federally endangered species, *Stephanomeria malheurensis*. Finally, the Hieraciinae has two *Hieracium* species formerly listed as Category 2, *H. pringlei* and *H. robinsonii*. We substituted a more common member from each genus (except *Microseris*) and tested *Hieracium albertinum*, *Lygodesmia juncea*, *Prenanthes sagittata*, and *Stephanomeria tenuifolia* along with several additional Lactucinae species: *Lactuca serriola*, *Lactuca sativa*, *Lactuca tartarica* var. *pulchella*, and *Lactuca virosa* (Table 3). For *Microseris*, we substituted the closely related Microseridinae species, *Agoseris aurantiaca*, as we were unable to locate a *Microseris* species at the time of testing. We also tested at least one species from each of the two other subtribes native to the continental Untied States which are in the same tribe as rush skeletonweed but do not contain species of concern; as well as one species from a subtribe with introduced species and another species from an unassigned genus. Because there was no larval feeding or development on any of these species (other than the small tube on *Lygodesmia juncea* described above), we do not believe *B. gilveolella* presents any risk to any listed or non-listed taxa other than *Chondrilla* species. We believe our studies combined with those of Caresche and Wapshere (1975) on a total of 37 Asteraceae species, present solid evidence that *B. gilveolella* poses little threat to any listed or non-listed Asteraceae species.

Table 3. TES species in the Lactuceae tribe and their substitute species for host specificity tests.

Subtribe	TES Species	Status[a]	Substitute Species
Crepidinae	*Taraxacum californicum*	E	*Taraxacum eriophorum*
	Taraxacum carneocoloratum	S	*Taraxacum laevigatum*
			Taraxacum officinale
			T. officinale ssp. *ceratophorum*
Hieraciinae	*Hieracium pringlei*	S	*Hieracium albertinum*
	Hieracium robinsonii	S	"
Lactucinae	*Prenanthes barbata*	S	*Prenanthes sagittata*
	Prenanthes boottii	S	"
Microseridinae	*Microseris decipens*	S	*Agoseris aurantiaca*
	Microseris howellii	S	"
Stephanomeriinae	*Lygodesmia dolorensis*	S	*Lygodesmia juncea*
	Stephanomeria blairii	S	*Stepahomeria tenuifolia*
	Stephanomeria malheurensis	E	"

[a] E = Endangered; S = Sensitive species formerly listed as Category 2 by the USFWS.

Potential impact on plant populations –

Larvae of *B. gilveolella* feed on the outer cortical portion of the rootstock, cutting cortical vessels and interrupting the flow of nutrients. The eventual damage to the root and plant is dependent upon the size of the root relative to the number of larvae feeding. Thin roots may be substantially cut by larval feeding, resulting in the death of the plant; whereas, larger plants are more tolerant of feeding. Caresche and Wapshere (1975) observed that in Greece one or two larvae were usually observed per root, whereas at more heavily infested sites, three to four larvae were more common - with up to ten larvae found on some large roots. Larval feeding may, as observed by Littlefield et al., create entry wounds for root rotting fungi, such as *Fusarium*, to infect plants. Such infection may also result in the death of the plant and may augment the decline in plant populations in suitable environments. Caresche and Wapshere (1975) speculated that heavy infestations of *B. gilveolella* would reduce the number of younger, thin rooted rush skeletonweed to create stands that would consist primarily of older, larger, more thick-rooted plants. Should this be the case, *B. gilveolella* may be more effective in newly formed infestations of the weed or on the periphery of well-established infestations where plants tend to be younger and smaller.

Habitat and climatic suitability –

The success of the moth in North America will be dependent upon meeting its ecological requirements. Studies in Greece by Caresche and Wapshere (1975) indicate that the moth does best in habitats with sandy, friable soils that are not compacted by grazing or other human manipulations, or those that lack heavy ground cover. Also from this study it appears that the moth is ill adapted to hot Mediterranean type climates with winter rainfall. Based upon these observations, we speculate that in North America the moth may be better suited for rush skeletonweed infestations found in Idaho, Montana, and eastern Washington and Oregon where the climate and soils are better suited.

IV. PROTOCOL FOR RELEASING THE AGENT

Geographical or Host Source:
The moth will be collected from rush skeletonweed in northern Greece. Collections are to be made from specific locations from which the original moth populations were collected for host specificity testing.

Method to Ensure Proper Identification:
Prior to field release, moths will be mounted and sent to taxonomists for identification. Adults of *B. gilveolella* and another root-boring moth, *Ena wertheimsteini*, are easily distinguishable; thus, no identification problems are anticipated in this regard. Screening procedures listed below will further prevent contamination.

Protocol to Ensure the Absence of Natural Enemies:
Infested roots will be received from Europe and will be placed on moist vermiculite in 10x20 propagation trays. Trays will be covered with domed lids, vented at either end and then placed in isolation cages to contain any unwanted organisms. The trays will be inspected for adult emergence. Parasites and other unwanted organisms will be removed, and destroyed. Voucher specimens of these will be kept. Larvae and adults will be sampled and inspected for the presence of pathogens. Additional rearing in quarantine may be required to eliminate pathogens, if present.

Impact of Other Management Practices:
Sites with minimal disturbance (i.e., from grazing, spraying, mowing, etc.) will be chosen.

Specific Location of Rearing Facility:
Quarantine handling and rearing will be conducted at the Insect Quarantine Laboratory, Montana State University, Bozeman.

Intended Sites for Initial Release:
Releases are proposed for selected sites in Boise and Kootenai Counties, located in central and northern Idaho; as well as an insectary site at Montana State University, Bozeman, Montana. The number of sites selected will be dependent upon the number of moths or moth eggs available.

Releases will be maintained and monitored by university researchers, county weed supervisors and/or federal land managers.

Number of Moths to be Released:
Moths will be released either as larvae or as adults. Approximately one hundred newly hatched larvae or 25 to 50 adult moths per site will be released, although actual numbers will be dependent upon availability of adult moths or eggs.

Timing of Releases:
Insectary site releases will be made in early to mid summer, depending upon moth development in Greece.

Release Methods:
Bradyrrhoa gilveolella will be held under quarantine at the Insect Quarantine Lab, Montana State University, Bozeman and screened for parasites and pathogens and confirmation of the species before being released. Methods for release will consist of inoculating plants with newly hatched larvae, or by releasing adult moths into field cages. Root samples will be taken in the fall and early spring to determine the presence of larvae on the roots. Releases will be caged the following year to contain emerging moths. The initial release of the moth will be conducted by quarantine personnel from Montana State University, assisted by other university researchers, local county weed supervisors, and/or federal land managers. The monitoring and maintenance of the site(s) will be by university researchers, local county weed supervisors, or federal land managers.

V. POTENTIAL ENVIRONMENTAL IMPACTS

Human Impacts:

1. Recreational: With less rush skeletonweed, fishing, hunting, and water/game activities will be more pleasurable.

2. Aesthetics: Eliminating or reducing rush skeletonweed will increase the diversity of grasses and wildflowers in infested areas.

3. Health Risks: *Bradyrrhoa gilveolella* is not known to pose a risk to human health, although wing scales may irritate the respiratory system if inhaled in quantity (especially if aspirating adult moths for collection). Some people may have mild allergic reactions to insect parts or frass material. These problems are usually associated with rearing facilities or handling of the insect.

Potential Economic Impacts:

The goal for biological control of rush skeletonweed is that a complement of agents will control the spread and reduce the density of rush skeletonweed, in all of the ecological niches in which it grows, to the extent that the application of herbicides or other control measures would no longer be necessary. The moth, *B. gilveolella* is one complement of this biological control program. With successful biological control, millions of dollars would be saved in direct management costs. Additional resources would be preserved as a result of avoiding side effects due to herbicide application. Native vegetation provides better forage for wildlife, and functions more efficiently at preventing soil erosion. A greater vegetative diversity supports a wider array of arthropods and vertebrates. Each of these have monetary equivalents which are difficult to accurately estimate but are evident — especially when managing

National Forests, Parks, or FWS lands for native vegetation, or rangelands for domestic forage, or recreation areas suitable for human enjoyment and education.

By controlling rush skeletonweed with biocontrol agents, less will be invested in the purchase and application of chemicals while simultaneously continuing to increase land values, the profits for hay and grazing operations, and the quality of soil and water resources. (Also refer to Target Weed Information 4. Economic Losses)

Plant Impacts:

Because *B. gilveolella* larvae feed exclusively on rush skeletonweed, no significant impacts are expected for non-target plants. The risk to native Asteraceae is low for reasons explained in the Discussion Section above. Indirect effects of successful biological control of rush skeletonweed by the moth would be the increase of plant biodiversity and the protection on non-target plants from the impacts associated with the use on nonselective herbicides.

Nonplant Impacts:

No direct, nonplant impacts are anticipated with the release and establishment of *B. gilveolella*. No significant interactions with other organisms are known to exist with *B. gilveolella* or with its host in non-native habitats. Indirect effects of successful biological control of rush skeletonweed by the moth would be the increase of biodiversity of the general ecosystem, the reduced potential for soil erosion and the protection of non-target organisms from the impacts associated with the use of nonselective herbicides.

Preventive/ Mitigating Methods:

No undesirable effects are anticipated, however, moth larvae may be eliminated by destroying their host plants by applications of herbicides, or by cultivation.

Abiotic/Edaphic Effects:

No significant effects to air resources are expected. The effect on water-soil systems should be positive for the reason that rush skeletonweed plants are less effective at preventing soil erosion than the native vegetation they replace. (Also refer to Target Weed Information - 9. Effect on ecosystem functions and ecological relationships)

Petitioner's Perspective of Risk:

From the results of the host specificity tests conducted in Europe and the United States, in combination with host records of the moth, we conclude that the Greek population of *B. gilveolella* is restricted to the genus *Chondrilla*. In the laboratory studies conducted in Montana, no feeding whatsoever was observed on any closely related plant species. Although a limited number of *Crepis* and *Taraxacum* were tested, we feel confident that the lack of feeding by larval *B. gilveolella* is indicative of the remaining members of those plant genera. Therefore we do not anticipate any feeding on non-target plants. In addition there is little risk in the release of *B. gilveolella* due to taxonomic confusion with other associated moths.

When considering the economic and ecological costs associated with rush skeletonweed infestations, unforeseen risks to non-target vegetation due the introduction of the moth are far out-weighed. Therefore we recommend its release.

Outcome of No Action:

Without releasing this moth the other biological control agents already released will continue to function. However, this root-feeding moth is able to fill specific niches where other agents are less capable. Without the moth's release and establishment these niches will not be covered as completely, allowing rush skeletonweed to persist and spread into new areas.

Conclusion:

In view of the field observations and laboratory studies, we feel that *B. gilveolella* can be a valuable agent for controlling rush skeletonweed. The moth is capable of inflicting severe damage to rush skeletonweed roots, and would be host-specific to *Chondrilla juncea* in North America. We recommend its release.

VI. ACKNOWLEDGEMENTS

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APPENDIX I. Rationale for the selection of test plants.

This section provides rationale as to the selection of plants for host specificity testing of *Bradyrrhoa gilveolella*, a possible biological control agent of *Chondrilla juncea* L. in North America. The list is based on the strategy in Wapshere (1974), *A Strategy for Evaluating the Safety of Organisms for Biological Weed Control*, published in Ann. Appl. Biol. 201-211. The strategy is based on the phylogenetic approach, where closely related species are theorized to be at greater risk of attack than are distantly related species. Included in this rationale are some species tested by Caresche and Wapshere (1975). Not included are various economic plant species (see Table 1), that are not associated with the phylogenic approach which we have taken.

Category 1:

Genetic types of *Chondrilla juncea* L. (varieties, races, forms, genotypes, apomicts, etc.).

Chondrilla juncea L. can exhibit great variation as evidenced by the morphologically distinct forms recognized in Europe. We tested five accessions representing three probable "biotypes":"Washington late-flowering", "Washington early-flowering", and "Banks". Rush skeletonweed collections were made from Pullman, Whitman County, Washington ("Washington late-flowering" – hereby designated WLF) (used as control plants); Placer and Nevada Counties California ("Washington late-flowering"); Liberty Lake, Spokane County, Washington ("Washington early-flowering"); and Banks, Boise County, Idaho ("Banks"). These locations were selected based on differences in plants noted by G. Markin and J. Littlefield. Given the current genetic knowledge available for *Chondrilla juncea* (Hasan et al. 1995), we believe that by testing plants from these locations, we will be testing all *Chondrilla juncea* "biotypes" currently reported in the western United States. However, as additional genetic knowledge becomes available, these biotypes should be reviewed. Since biotypes may differ in their susceptibilities to very host specific biocontrol agents, testing of these three recognized biotypes may not enable researchers to make inferences about susceptibility of any additional genotypes that are identified in the future.

Testing by Caresche and Wapshere (1975) included several European forms or "biotypes" of rush skeletonweed in addition to the three Australian forms.

Category 2:

North American species in the same genus as *Chondrilla juncea* L., divided by subgenera (if applicable), including economically and environmentally important plants.

Although there are about 25 species of *Chondrilla* worldwide, there are no *Chondrilla* native to North America (Bremer 1994). Because no *Chondrilla* species, other than *Chondrilla juncea*, have been reported in North America (Kartesz and Kartesz 1980, USDA-NRCS 1999), we did not test additional *Chondrilla* species.

Category 3:

North American species in other genera in the Asteraceae family, divided by subtribe, tribe, and subfamily, including economically and environmentally important plants.

The Asteraceae is the largest plant family and is divided into 3 subfamilies, 17 tribes, and numerous subtribes. In developing our list of plants to be tested, we followed the recent reclassification of the Asteraceae by Bremer (1994). Bremer treated the Asteraceae from a cladistic perspective, based mainly on morphological characteristics. He grouped taxa according to shared common ancestry and tried to rewrite taxa to be monophyletic (include all descendants from a common ancestor). His cladograms may be interpreted as phylogenetic trees or, more precisely, phylogenetic hypotheses.

Chondrilla juncea L. belongs to the Asterales order, Asteraceae family, Cichorioideae (= Lactucoideae) subfamily, Lactuceae (formerly named Cichorieae) tribe, Crepidinae subtribe, and *Chondrilla* genus.

Species in the same subtribe (Crepidinae) as *Chondrilla juncea* L.:

Two genera in the Crepidinae subtribe (*Crepis* and *Taraxacum*) contain species native to the United States (Bremer 1994).

Crepis is an important genus to test because of the number of native species in the Western United States (USDA-NRCS 1999) and their biochemical similarity with *Chondrilla*. *Crepis* and *Chondrilla* are similar in phenolic compounds (which have a high chemotaxonomic value) in that at least some species of both *Crepis* and *Chondrilla* contain quercitin and lack apigenin derivatives (Mañez et al. 1994, Rees and Harborne 1984). Rather than test any introduced species, we tested 4 native species of *Crepis*. The four species tested were *Crepis acuminata* Nutt., *Crepis atribarba* Heller, *Crepis runcinata* (James) Torr. & Gray, and *Crepis elegans* Hook.. These species were available at the time of testing.

Taraxacum is an important genus to test because of the presence of native species in the Western United States (USDA-NRCS 1999), some of which are threatened, endangered, or sensitive (see Category 4). We tested four *Taraxacum* species. The introduced *T. officinale* G. H. Weber ex. Wiggers was suggested because it was attacked by several candidate biological control agents in previous tests (Caresche *et al.* 1974, Caresche and Wapshere, 1974, Caresche and Wapshere 1975a, Hasan and Wapshere 1977) and was readily available. Similarly, the introduced species *Taraxacum laevigatum* (Willd.) DC was tested because of its availability. We also selected for testing two native species found in the Western United States: *Taraxacum eriophorum* Rydb. and *T. officinale* ssp. *ceratophorum* (Ledeb.) Schinz ex. Thellung (=*Taraxacum ceratophorum* Ledeb)(USDA-NRCS 1999). We believe that by testing these four *Taraxacum* species, we obtained sufficient information to infer if any non-tested *Taraxacum* species would be at risk.

Three other genera in the Crepidinae subtribe (*Ixeris*, *Lapsana*, and *Youngia*) are not native but are listed as present in the continental United States (Bremer, 1994, Kartesz and Kartesz, 1980, USDA-NRCS 1999). Although testing species from these genera would provide further information on a potential biological control agent's host range, we did not test any species from these genera. Since all species

from these genera are introduced weeds in the United States, attack by a potential biological control agent should not result in any conflicts of interest.

Species in different subtribes in the same tribe (Lactuceae):

The Lactuceae are one of the best phylogenetically known tribes of the Asteraceae, and are well supported as a distinct monophyletic group. Subtribal and generic classifications are more defined in this tribe than in most other tribes (Bremer 1994). The Lactuceae are set apart from other Asteraceae by a ligulate capitula, milky latex, and absence of oil ducts (except in *Scolymus*), although none of these characteristics are restricted to the Lactuceae alone (Mañez *et al.* 1994). The Lactuceae also differ phytochemically in that their sesquiterpene lactones are predominately guaianolides whereas other Asteraceae subfamilies contain plants with considerable portions of other sesquiterpene lactones (Wapshere 1983). Unlike most other Asteraceae, the Lactuceae possess either no or low levels of polyacetylene compounds (Sorensen 1977). Besides the Crepidinae, there are six Lactuceae subtribes that contain species native to the continental United States (Lactucinae, Sonchinae, Microseridinae, Stephanomeriinae, Malcothrinae, Hieraciinae); three subtribes with some introduced species (Catananchinae, Hypochaeridinae = Leontodontinae, and Scorzonerinae); and two introduced genera (*Cichorium* and *Scolymus*) that have not been assigned to a subtribe (Bremer 1994).

Of these subtribes, the Hypochaeridinae may be most closely related to the Crepidinae. Like *Chondrilla*, most Hypochaeridinae species contain quercitin and lack apigenin (Giner *et al.* 1993; Mañez *et al.* 1994). Quercitin or its glycosides have been identified in the genera *Hypochaeris*, *Leontodon, Picris*, and *Urospermum* (Giner *et al.* 1993). *Hypochaeris glabra* L. and *Picris echioides* L. were attacked by biological control agents in previous tests (Caresche *et al.* 1974). Both are listed as present in North America (Kartesz and Kartesz 1980) but are weedy species (Bremer 1994). We did not test any species from the Hypochaeridinae, as this subtribe is not native and contains no commercially important introduced species in North America.

Chondrilla has also been chemotaxonomically linked with the genera *Sonchus* and *Lactuca* because these genera contain quercitin and its glycosides (Mañez *et al.* 1994). Because of the economic importance of *Lactuca sativa* L. (lettuce), we tested three varieties of the four basic lettuce types: butterhead, crisphead, looseleaf, and romaine. We also tested *Lactuca virosa* L. which, along with species of *Chondrilla* and *Hieracium*, has been shown to contain the benzoic derivative, protocatechuic acid (Mañez *et al.* 1994). In addition, we tested *Lactuca tartarica* var. *pulchella* (Pursh.) Breitung, a native lettuce found in the Western United States. As there are no native *Sonchus* in the Western United States (Hickman 1996, Hitchcock and Cronquist 1973, USDA-NRCS 1999), we tested *Sonchus oleraceus* L. and *S. uliginosus* L.. Caresche and Wasphere (1975) tested both *S. oleraceus* and *S. arvensis* L.

Quercitin or its glycosides have also been identified as common in *Scorzonera* and *Tragopogon* and present in other genera including *Agoseris*, *Cichorium*, *Krigia*, *Microseris*, and *Scolymus*, (Giner *et al.* 1993, Rees and Harborne 1984). *Scorzonera hispanica* L., *Tragopogon porrifolius* L., *Cichorium intybus* L., *Cichorium endivia* L., and *Scolymus hispanicus* L. were attacked by biological control agents in previous tests (Caresche *et al.* 1974, Hasan, 1978, Hasan and Wapshere 1977). We tested *Cichorium intybus* L., and *Agoseris aurantiaca* (Hook.) Greene. Caresche and Wasphere (1975) tested both *C. intybus*, and *C. endivia.* We did not test any species of *Scolymus*, *Scorzonera*, or *Tragopogon*

as these genera are not native to North America (Bremer 1994), although Caresche and Wasphere (1975) tested *Scorzonera hispanica*. While *Krigia* is native to the Central and Southern United States, we did not test this genus since no species are found in the Western United States (Bremer 1994, USDA-NRCS 1999). Furthermore, we believe that by testing *Agoseris* and *Cichorium*, along with the two quercitin-containing species proposed in Category 6, we obtained sufficient information to infer if *Krigia* or any other non-tested quercitin-containing genera/species would be at risk. We also tested *Catananche caerula* L., which is an ornamental species.

By testing these species, combined with those in Category 4, we have tested at least one species from each of the six subtribes native to the continental United States, one species from a subtribe with introduced species (Catananchinae), and one species from an unassigned genus (*Cichorium*). We believe that by testing these species, we obtained sufficient information to infer if plants in Lactuceae subtribes, other than the Crepidinae, would be at risk.

Species in different tribes in the same subfamily (Cichorioideae):

According to Bremer (1994), in the Cichorioideae subfamily there are three other tribes, besides Lactuceae, with natives in North America (Cardueae/Cynareae, Mutisieae, and Vernonieae) plus a tribe with some introduced ornamentals (Arctoteae). The Mutisieae and the Vernonieae are believed to be closely related to the Lactuceae (Mañez *et al.* 1994, Tomb 1977). Like the Lactuceae, several species of Mutisieae and *Stokesia* of the Vernonieae have ligulate corollas (Tomb 1977). We tested *Gerbera jamesoni* Bolus. (an introduced ornamental) and *Stokesia laevis* (Hill) Greene (a native ornamental). We also tested *Gazania splendens* E. G. & A. Henderson (an introduced ornamental) of the Arctoteae as *Gazania* species are similar to the Lactuceae in producing latex (Tomb 1977). *Gundelia*, *Berardia*, and *Warionia* also share similarities with the Lactuceae, but were not tested because there are no native or economic species in United States (Bremer 1994, Mabry and Bohlmann 1977).

From the Cardueae/Cynareae, we tested *Cirsium undulatum* (Nutt.) Spreng. (see Category 4). Caresche and Wasphere (1975) tested the economically important species, *Carthamus tinctorius* L. (safflower) and *Cynara scolymus* L. (globe artichoke), from this tribe.

We have tested at least one species from each of the three tribes native to the continental United States and one species from a tribe with introduced species (Arctoteae). We believe that by testing these species, we obtained sufficient information to infer as to whether any plants in the Cichorioideae subfamily would be at risk.

Species in different subfamilies in the same family (Asteraceae):

Besides the Cichorioideae, there are two additional subfamilies (Barnadesioideae and Asteroideae) in the Asteraceae family (Bremer 1994).

The Barnadesioideae comprise nine genera and are exclusively South American. Because there are no species native to North America (Bremer 1994), we did not include any Barnadesioideae for testing.

The Asteroideae include the majority of the Asteraceae and comprise eight tribes with species native to the United States (Anthemideae, Astereae, Eupatorieae, Gnaphalieae, Helenieae, Heliantheae,

Plucheeae, and Senecioneae), two additional tribes with ornamental species (Calenduleae and Inuleae), and eight genera unassigned to a tribe (Bremer 1994). Although acetylenes are relatively rare in the Lactuceae, those which are present belong to a special group which may link the Lactuceae chemotaxonomically with the Astereae and the Anthemideae (Mabry and Bohlmann 1977).

Caresche and Wasphere (1975) tested species representing four tribes of Asteraceae: Anthemideae - *Chrysanthemum indicum* L., *Chrysanthemum leucanthemum* L.; Calenduleae - *Calendula* sp.; Helenieae - *Tagetes erecta* L.; and Heliantheae - *Dahlia* sp., *Zinnia* sp., *Helianthus annuus* L., *Helianthus tuberosus* L..

Because at least one species from three of the eight Asteroideae tribes native to the continental United States was tested, we believe that we have sufficient information to infer if *B. gilveolella* is likely to attack plants in the Asteraceae family outside the Cichorioideae subfamily.

Category 4:

Threatened, endangered, and sensitive species in the Asteraceae family, divided by subgenus, genus, subfamily, and tribe (see Table 2).

Species in the same subtribe (Crepidinae) as Chondrilla juncea:

Two *Taraxacum* species are listed as threatened, endangered, or sensitive in the continental United States. *Taraxacum californicum* Munz. & Johnston is found in California and is currently listed as endangered (USFWS 1999). We did not test *T. californicum* since the seeds we were able to obtain were not viable and to obtain sufficient viable seed or plant material for host specificity tests could further jeopardize the species. *Taraxacum carneocoloratum* A. Nels, historically found in Alaska and the Yukon (outside the current range of *Chondrilla juncea*), was formerly listed as a Category 2[1] species (USFWS 1993). From Category 3, we tested four *Taraxacum* taxa. We believe that by testing four *Taraxacum* taxa, we obtained sufficient information to infer if *T. californicum* or *Taraxacum carneocoloratum* would be at risk of attack.

Species in different subtribes in the same tribe (Lactuceae):

Four of the subtribes have threatened, endangered, or sensitive species in the continental United States (USFWS 1993, USFWS 1996). The Lactucinae has two species of *Prenanthes* formerly listed as Category 2[1] (*P. barbata* (Torr. & Gray) Milstead and *P. boottii* (DC.) Gray). The Microseridinae has two species of *Microseris* formerly listed as Category 2[1] (*M. decipens* Chambers and *M. howellii* Gray). The Stephanomeriinae formerly had two Category 2[1] species: *Lygodesmia doloresensis* S. Tomb and *Stephanomeria blairii* Munz. & Johnston; and one federally endangered species: *Stephanomeria malheurensis* Gottlieb. The Hieraciinae formerly had two Category 2[1] Hieracium species (*H. pringlei* Gray and *H. robinsonii* (Zahn) Fern.).

Because obtaining seeds and/or plant material of threatened, endangered, and sensitive species can be difficult and may further decimate populations, we decided not to test the threatened, endangered, or sensitive species. Instead, we substituted a more common member from each genus. We tested

Hieracium albertinum Farr = *Hieracium scouleri* var. *albertinum* Hook., *Lygodesmia juncea* (Pursh) D. Don ex Hook., *Prenanthes sagittata* (Gray) A. Nels., and *Stephanomeria tenuifolia* (Torr.) Hall.

Species in different tribes in the same subfamily (Cichorioideae):

The Cardueae/Cynareae has several *Cirsium* species listed as threatened, endangered, or sensitive in the continental United States (USFWS 1993, USFWS 1996). *C. fontinale* var. *fontinale* (Greene) Jeps. and *C. fontinale* var. *obispoense* J. T. Howell are federally endangered. *C. pitcheri* (Torr. ex Eat.) Torr. & Gray and *C. vinaceum* Woot. & Standl. are federally threatened. *C. hydrophilum* var. *hydrophilum* (Greene) Jeps. is proposed for listing as endangered. The Fish and Wildlife Service has sufficient information on *C. loncholepis* Petrak and *C. rhothophilum* Blake to propose them for listing as threatened or endangered. *C. crassicaule* (Greene) Jeps., *C. fontinale* var. *campylon* (H. K. Sharsm.) Pilz ex Keil & C. Turner, *C. hillii* (Canby) Fern., *C. hydrophilum* var. *vaseyi* (Gray) J. T. Howell, *C. longistylum* Moore & Frankton, *C. occidentale* var. *compactum* Hoov., *C. ownbeyi* Welsh, *C. parryi* ssp. *mogollonicum* C. Schaack & G. Goodwin, and *C. virginense* Welsh were formerly listed as Category 2[1] species.

Because obtaining seeds and/or plant material of threatened, endangered, and sensitive species can be difficult and can further decimate populations, we decided not to test the threatened, endangered, or sensitive species. Instead, we tested *Cirsium undulatum* (Nutt.) Spreng., a more common member from this genus and phylogenetically related to *C. pitcheri*.

Species in different subfamilies in the same family (Asteraceae):

At this level, we do not plan to test representatives from genera with threatened, endangered, or sensitive individuals. With over 20 genera with federally listed threatened or endangered species and over 70 additional genera with sensitive species in the continental United States, the number of species we would need to test is prohibitively large. Instead, we concentrated on testing species which were attacked by other biological control agents in previous tests, economically important species, and species which are readily available (see Category 3). We believe that by testing these species, we were able to infer if *B. gilveolella* is likely to attack any threatened, endangered, or sensitive species, in the Asteraceae family outside the Cichorioideae subfamily.

Category 5:

North American species in other families in the Asterales order that have some phylogenetic, morphological, or biochemical relationship to the target weed, including economically and environmentally important plants.

Neither Cronquist nor Dahlgren list any families other than the Asteraceae in the Asterales order; however, Thorne lists the Calyceraceae as belonging in the Asterales (see Appendix 7 of the proposed TAG Reviewer's Manual). Cronquist places the Calyceraceae in the Calycerales while Dahlgren places it in the Dipsacales. Since two out of three systems list the Calyceraceae in an order other than Asterales, we discuss this family in Category 6.

Category 6:

North American species in other orders that have some morphological or biochemical relationship to the target weed, including economically and environmentally important plants.

Species in other orders that are phylogenetically related to the Asteraceae:

The Asteraceae form such a well-defined group that they are sometimes considered systematically isolated. The most closely related families are generally considered to be the Calyceraceae, Campanulaceae *sensu lato*, and Goodeniaceae (Bremer 1994). The Calyceraceae contain six genera with about 60 species in southern South America (Bremer 1994). One species, *Acicarpha tribuloides* Juss., is listed as present in North America (Kartesz and Kartesz 1980) but was not selected for testing. The Campanulaceae *sensu lato* contain about 85 genera and more than 2200 species (Bremer 1994). There are about 23 genera and 290 species in the United States and Canada (Zomlefer 1994). The Campanulaceae have been shown to be chemotaxonomically linked to the Asteraceae (Mabry and Bohlmann 1977). The Goodeniaceae contain 12 genera and 400 predominately Australian species (Bremer 1994). One genus, *Scaevola*, has 2 species listed as present in the Central and Southern United States (USDA-NRCS 1999) but neither was selected for testing.

Species in other orders with biochemical characteristics in common with the Asteraceae:

The Lactuceae are characterized, in part, by their milky latex. However, more than 12,500 species in 900 genera and 20 families have been identified worldwide that produce latex (Metcalfe 1967). Over 300 species in the United States have been shown to contain latex (Buchanan et al. 1978a). Caresche and Wapshere (1975) tested one of these latex-producing species: *Allium cepa* L..

The Apiaceae (= Umbellifereae) have been shown to be chemotaxonomically linked to the Asteraceae (Mabry and Bohlmann, 1977). Like the *Chondrilla*, some Apiaceae have been shown to contain quercitin (Crowden et al. 1969). Caresche and Wapshere (1975) tested *Pastinaca sativa* L. which contains quercitin and is economically important.

Vigna radiata (L.) R. Wilczek of the Fabaceae family was also tested because it contains an unusual compound, tartronic acid-2-caffeoyl-ester, which has been found in rush skeletonweed (Mañez *et al.* 1994).

Selected cultivated species in other orders:

Caresche and Wapshere (1975) tested several crop species in addition to the ones we have listed above. These included: *Beta vulgaris* L., *Phaseolus vulgaris* L., *Pisum sativum* L., and *Saccharum officinarum* L..

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.

Little is known regarding the host plants of *Bradyrrhoa* species. Host plant associations have been made for only two species, *B*. *gilveolella* and *B*. *lyratella*. Both species feed on members of the Lactuceae: *Chondrilla* spp. and *Andryala lyrata* (respective to the moth species). Members of the genus *Andryala* are not found in North America.

Perspective of Risk:

We believe that by testing these species we were able to infer the potential host range of *B*. *gilveolella*. Because of the importance of the Asteraceae family, both environmentally and economically, we believe any potential biological control agents which are not specific to the genus *Chondrilla* should either not be approved for release in the United States or receive a detailed risk-benefit analysis before being considered for approval.

Footnotes

[1]In 1993, the U. S. Fish and Wildlife Service assigned Category 2 status to taxa for which the Service had information indicating that proposing to list as threatened or endangered was possibly appropriate, but for which sufficient data on biological vulnerability and threat were not currently available to support proposed rules (USFWS, 1993). In 1996, the U. S. Fish and Wildlife Service completed an exhaustive review of the 1993 list. Listing of Category 2 species was discontinued although the Service remains concerned about and acknowledges the need for further biological and field study of these species (USFWS, 1996).

TABLE 4. List of plant species tested to determine the potential host range of *Bradyrrhoa gilveolella*, a **candidate biological control agents of *Chondrilla juncea* L. in the United States (including a partial listing of plants tested by Caresche and Wapshere 1975).**

Plant Species	Common Name	Origin[a]	Classification[b]	Life Cycle[c]	Root[d]
CATEGORY 1: Genetic types of *Chondrilla juncea* L.					
Chondrilla juncea (WLF)	rush skeletonweed	I	Crepidinae	P	T
Chondrilla juncea (Banks)	rush skeletonweed	I	Crepidinae	P	T
Chondrilla juncea (WEF)	rush skeletonweed	I	Crepidinae	P	T
Chondrilla juncea (WLF)	rush skeletonweed	I	Crepidinae	P	T
CATEGORY 2: Species in the same genus as *Chondrilla juncea* L.					
None in North America.					
CATEGORY 3: Species in the other genera in the Asteraceae family.					
Species in the same subtribe (Crepidinae) as *Chondrilla juncea* L.					
Crepis acuminata	longleaf hawksbeard	N	Crepidinae	P	T
Crepis atribarba	slender hawksbeard	N	Crepidinae	P	T
Crepis elegans	elegant hawksbeard	N	Crepidinae	P	T
Crepis runcinata	fiddle-leaf hawksbeard	N	Crepidinae	P	T
Taraxacum eriophorum	wool-bearing dandelion	N	Crepidinae	P	T
Taraxacum laevigatum	rock dandelion	I	Crepidinae	P	T
Taraxacum officinale	common dandelion	I	Crepidinae	P	T
Taraxacum officinale ssp. ceratophorum	horned dandelion	N	Crepidinae	P	T
Species in different subtribes in the same tribe (Lactuceae).					
Agoseris aurantiaca	orange agoseris	N	Microseridinae	P	T
Catananche caerulea	Cupid's dart	I	Catananchinae	P	F
Cichorium endivia	endive	I	Unassigned	P	T
Cichorium intybus	chicory	I	Unassigned	P	T
Hieracium scouleri	wooly-weed	N	Hieraciinae	P	R
Lactuca sativa (Var. 1)	iceberg lettuce	C	Lactucinae	A	T
Lactuca sativa (Var. 2)	Grand Rapids lettuce	C	Lactucinae	A	T
Lactuca sativa (Var. 3)	romaine lettuce	C	Lactucinae	A	T
Lactuca serriola	prickly lettuce	N	Lactucinae	P	C
Lactuca tartarica var. pulchella		I	Lactucinae	B	T
Lactuca virosa	bitter lettuce	I	Lactucinae	B	T
Lygodesmia juncea	rush-like skeletonweed	N	Stephanomeriinae	P	
Prenanthes sagittata	arrowleaf rattlesnakeroot	N	Lactucinae	P	
Scorzonia hispanica		I	Scornzonerinae		

Table 4. Continued.

Plant Species	Common Name	Origin[a]	Classification[b]	Life Cycle[c]	Root[d]
Stephanomeria tenuifolia	narrow-lvd. wirelettuce	N	Stephanomeriinae	P	
Sonchus arvensis	perennial sowthistle	I	Sonchinae	P	T/R
Sonchus oleraceus	common sowthistle	I	Sonchinae	A	T
Sonchus uliginosus	marsh sowthistle	I	Sonchinae	P	T

Species in different tribes in the same subfamily (Lactucoideae/Cichorioideae).

Carthamus tinctorius	safflower	I	Cardueae/Cynareae	A	
Cynara scolymus	artichoke	I	Cardueae/Cynareae	P	
Cirsium undulatum	wavy-leaf thistle	N	Cardueae/Cynareae	P	T/R
Gazania splendens	daybreak gazania	I	Arctoteae	P	F
Gerbera jamesoni	Transvaal daisy	I	Mutisieae	P	F
Stokesia laevis	Stokes Aster	N	Vernonieae	P	F

Species in different subfamilies in the same family (Asteraceae).

Calendula sp.	pot marigold	I	Calenduleae	A	
Chrysanthemum indicum	chrysanthemum	I	Anthemideae		
Chrysanthemum leucanthemum	oxeye-daisy	I	Anthemideae	P	T/R
Dahlia sp.		I	Heliantheae	P	
Helianthus annuus	sunflower	N	Heliantheae	A	T
Helianthus tuberosus	Jerusalem artichoke	N	Heliantheae	A	
Tagetes erecta	tall marigold	I	Helenieae	A	F
Zinnia sp.		I	Heliantheae	A	

CATEGORY 4: Threatened, endangered, or sensitive species in the Asteraceae family.

Related Species in the same subtribe (Crepidinae) as *Chondrilla juncea* L.

Taraxacum eriophorum	woolbearing dandelion	N	Crepidinae	P	T
Taraxacum laevigatum	rock dandelion	I	Crepidinae	P	T
Taraxacum officinale	common dandelion	I	Crepidinae	P	T
Taraxacum officinale ssp. ceratophorum	horned dandelion	N	Crepidinae	P	T

Table 4. Continued.

Plant Species	Common Name	Origin[a]	Classification[b]	Life Cycle[c]	Root[d]
Related Species in different subtribes in the same tribe (Lactuceae).					
Hieracium scouleri	wooly-weed	N	Hieraciinae	P	R
Lygodesmia juncea	rush-like skeletonweed	N	Stephanomeriinae	P	C
Prenanthes sagittata	arrowleaf rattlesnakeroot	N	Lactucinae	P	T
Stephanomeria tenuifolia	narrow-lvd. wirelettuce	N	Stephanomeriinae	P	T
Related Species in different tribes in the same subfamily (Lactucoideae/Cichorioideae).					
Cirsium undulatum	wavy-leaf thistle	N	Cardueae/Cynareae	P	T,R
Species in different subfamilies in the same family (Asteraceae).					
See Category 3.					
CATEGORY 5: Species in other families in the Asterales order.					
None.					
CATEGORY 6: Species in other orders that have some relationship to Chondrilla juncea L.					
Species in other orders with biochemical characteristics in common with the Asteraceae:					
Allium cepa	onion	I	Liliaceae	A,B	
Pastinaca sativa	parsnip	I	Apiaceae	A,B	T
Vigna radiata	mung bean	I	Fabaceae	A	
Selected cultivated species in other orders:					
Beta vulgaris	beet	I	Chenopodiaceae	A,B	T
Phaseolus vulgaris	french bean	N	Fabaceae	A	
Pisum sativum	garden pea	I	Fabaceae	A	
Saccharum officinarum	sugarcane	C	Gramineae	P	
Vicia faba	broad bean	I	Fabaceae	A	

CATEGORY 7: Species reported as attacked by the biological control agent or its close relatives

None tested

[a]Origin: C, Cultigen; I, Introduced; N, Native.
[b]Classification: Categories 1 through 4 list subtribe or tribe; Categories 5 through 7 list family.
[c]Life Cycle: A, Annual; B, Biennial; P, Perennial.
[d]Root: C, Deep-seeted creeping root; F, Fibrous; R, Rhizome; T, Taproot.