

# Biological control of invasive water primroses, *Ludwigia* spp., in the United States: A feasibility assessment

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## ABSTRACT

Exotic water primroses are aggressive invaders in both aquatic and riparian ecosystems worldwide. Water primrose [*Ludwigia hexapetala* (Hook. & Arn.) Zardini, Gu & P. H. Raven], floating primrose-willow [*Ludwigia peploides* (Kunth) P. H. Raven subsp. *peploides*], floating primrose-willow [*Ludwigia peploides* (Kunth) P. H. Raven subsp. *montevicensis* (Spreng.) P. H. Raven], Uruguay waterprimrose [*Ludwigia grandiflora* (Michx.) Greuter & Burdet], and the winged waterprimrose (*Ludwigia decurrens* Walter) have naturalized in aquatic ecosystems in the United States and are the focus of this study. The only control tools available to resource managers for suppression of *Ludwigia* spp. are physical and chemical methods, but these options are often limited in effectiveness and by costs and regulatory constraints. Biological control is an alternative that can be used alone or in combination with traditional methods. The purposes of this study were to explore the feasibility of a biological control program targeting problematic *Ludwigia* spp. in the United States and to propose a list of plant species for consideration during host range studies of candidate herbivores. A variety of native insects feed on *Ludwigia* spp. in the United States; however, most are generalists and have no appreciable influence on plant growth or fitness. Foreign exploration for natural enemies of *Ludwigia* spp. in South America suggests that a rich herbivore fauna is associated with the plants in their native range. Candidate agents must have section-level host specificity because several *Ludwigia* spp. are also native to the United States. Therefore, the plant test list is designed to distinguish herbivore host ranges based on the phylogenetic relationships of the test plants. For those *Ludwigia* spp. for which eradication may no longer be possible because the weed is regionally abundant, biological control may be the primary control option when traditional methods are not feasible.

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*Key words:* aquatic weed, host range, invasive species, primrose-willow, weed management.

## INTRODUCTION

Freshwater aquatic ecosystems grow increasingly susceptible to invasions by exotic plants due in part to limited regulations on importation pathways and eutrophication of waterways (Lodge et al. 1998, Gallardo et al. 2016). Among the most problematic invasive aquatic plants are select water primroses (*Ludwigia* L. spp.), which occur in both aquatic and riparian ecosystems in many geographic regions (EPPO 2011, Thouvenot et al. 2013). They are considered among the 200 most-aggressive weeds worldwide (Cronk and Fuller 1995). The genus exhibits a complex biogeographic pattern, with 10 sections endemic or centered in South America (40 spp.), three in North America (23 spp.), five in Africa (7 spp.), three in Asia (3 spp.), and two not clearly centered in a single continent (10 spp.). *Ludwigia* spp. is a large and nearly cosmopolitan monophyletic genus of wetland plant species within the Onagraceae family, currently classified into 23 sections with 88 taxa, including 83 species (Wagner et al. 2007, Pesamosca and Boldrini 2015, Liu et al. 2017). Most (80%) are New World species distributed in the Americas, although the genus is pantropical with some (largely naturalized) representation in temperate Europe, Africa, and Eurasia (Wagner et al. 2007). *Ludwigia* spp. is the center of origin and basal lineage of the entire Onagraceae family (thus, a sister group to the remainder of the genera in the family), being the first genus to branch from the common ancestor of Onagraceae between 80 and 93 million yr ago (Wagner et al. 2007). The divergence likely occurred in South America because the sections of *Ludwigia* spp. with the most plesiomorphic features center there.

The popularity of ornamental plants in the 19th century in the United States resulted in the introduction and naturalization of many pest plants (Reichard and White 2000). Exotic *Ludwigia* taxa are popular aquarium and water garden plants, which have subsequently naturalized and become significant weeds in wetland habitats (Dandelot et al. 2005b, Wagner et al. 2007, Okada et al. 2009). In the United States, exotic *Ludwigia* taxa from the largely aquatic *Ludwigia* sect. *Jussiaea* (Hoch et al. 2015) are considered invasive in aquatic systems of the Atlantic, Gulf Coast, and Pacific Northwest coastal states (Hoch and Grewell 2012, Grewell et al. 2016a): water primrose [*Ludwigia hexapetala* (Hook. & Arn.)

Zardini, Gu, & P. H. Raven], floating primrose-willow [*Ludwigia peploides* (Kunth) P. H. Raven subsp. *peploides*], floating primrose-willow [*Ludwigia peploides* (Kunth) P. H. Raven subsp. *montevideensis* (Spreng.) P. H. Raven], and Uruguay waterprimrose [*Ludwigia grandiflora* (Michx.) Greuter & Burdet]. A fifth invasive species, the erect, annual winged waterprimrose (*Ludwigia decurrens* Walter), from the *Ludwigia* sect. *Pterocaulon*, is invasive in rice (*Oryza sativa* L.) fields of California and southeastern states.

The exotic *Ludwigia* taxa in the United States are perennial forbs that readily colonize damp terrestrial to aquatic habitats. At the water's edge, the emergent plants produce buoyant, leafy shoots that can form mats across the water's surface (Grewell et al. 2016a). Generally, throughout their invaded ranges, these exotic *Ludwigia* taxa are tolerant of a wide range of environmental conditions and readily invade habitats of varying hydrological and climatic conditions (Thouvenot et al. 2013, Sarat et al. 2015). Plants are reported from wetlands; slow-flowing rivers, along edges of lakes and reservoirs, from gravel and mud river banks, ponds, irrigation ditches, wet meadows, and in the shallow waters of floodplains in the United States (Wagner et al. 2007, Hoch and Grewell 2012, Grewell et al. 2016a). Buoyant asexual fragments and seed capsules can quickly disperse with flowing water and colonize throughout watersheds (Okada et al. 2009, Skaer Thomason et al. 2018b).

#### IDENTIFICATION, ORIGIN, AND GEOGRAPHIC DISTRIBUTION

It is widely accepted that the genus *Ludwigia* originated in southern South America, the center of diversity for the genus, both in number of species and in intraspecific morphological variability (Raven and Tai 1979, Zardini and Raven 1992). Because the 13 polyploid taxa in sect. *Jussiaea* are highly polymorphic and morphological distinctions among closely related *Ludwigia* species are often not well defined, they are difficult to identify (Zardini et al. 1991b). The genus has undergone extensive taxonomic revisions since Raven (1963a). These revisions are largely based on morphology and the ploidy levels of the species (Wagner et al. 2007, Hoch et al. 2015), with few molecular studies that have been limited in scope to a particular region. As ploidy levels of *Ludwigia* vary by species, the target taxa and potential hybrids can be distinguished from one another by counting chromosome numbers (Zardini et al. 1991a,b). Ploidy levels refer to the number of sets of chromosomes in the nucleus of a biological cell. The base number of chromosomes in *Ludwigia* is  $n = 8$ ; *L. peploides* and *L. decurrens* are diploid ( $2n = 16$  chromosomes), *L. grandiflora* is hexaploid ( $2n = 48$  chromosomes), and *L. hexapetala* is decaploid ( $2n = 80$  chromosomes).

The morphology of the target taxa shows great phenotypic plasticity in response to environmental conditions (e.g., whether the plants are growing on moist soil or floating in water), and published taxonomic keys reflect characteristics of the region in which the study took place. Morphological variation also varies with life stage, changing through the year (e.g., variation in overall size, leaf size, stem vestiture, and other characteristics). Nearly all species of the sect.

*Jussiaea* can form vigorous interspecific hybrids, and at least one naturally occurring hybrid has been documented in Asia (Zardini et al. 1991b). Octoploid hybrids ( $2n = 64$ ) between *L. hexapetala* and *L. grandiflora* with intermediate morphology were confirmed in southern Brazil (Zardini et al. 1991a,b). Recently, Grewell and Gaskin (unpub. data) discovered octoploid *Ludwigia* hybrids naturalized in Florida, and Okada et al. (unpub. data) suspect recent hybridization. To further compound problems with identification, conflicting identifications that show large divergences in the choice of diagnostic criteria have been presented in the United States (Munz 1942, Zardini et al. 1991a, Nesom and Kartesz 2000, Hoch et al. 2015), and in Europe (Dandelot 2005b and references therein). Therefore, to properly differentiate species, a combination of methods, including field observations of growth characteristics, morphological evaluation of fresh specimens, and chromosome number counts, are employed (Grewell et al. 2016a). Proposed taxonomic revisions that have not been widely accepted relied solely on morphological evaluation of limited herbaria specimens with incomplete information on chromosome numbers, genotype, and high potential for hybrids (e.g., Nesom and Kartesz 2000). Detailed descriptions of the target taxa are published in journals (Zardini et al. 1991a,b), and in several taxonomic reference books specific to a particular state or region (e.g., Hoch and Grewell 2012).

In an extensive revised classification of the Onagraceae family, four of the five focal taxa in this study described from the monophyletic, aquatic *Ludwigia* sect. *Jussiaea* were considered likely exotic invaders in the United States (Wagner et al. 2007). However, in several taxonomic treatments, some long-established *Ludwigia* taxa were thought to be native to the United States. For example, native status was attributed to *L. peploides* subsp. *peploides* in California (Hickman 1993). The taxa's amphitropical and disjunct distribution from the South American native range (Raven 1963a,b) and recent phylogenetic results (Liu et al. 2017) have now provided convincing support for recognition of the South American origin for the entire sect. *Jussiaea*, including all *Ludwigia peploides* taxa in the United States. The native distribution of target taxa from this section centers in southern South America (Brazil, Uruguay, Paraguay, and/or Argentina) and Central America for *L. grandiflora*. In addition, creeping water primrose [*Ludwigia peploides* (Kunth) P. H. Raven subsp. *glabrescens* (Kuntze) P. H. Raven] has long been considered native in the eastern to midwestern United States, but it has been observed in South America, and molecular data now ascribe the origin to South America. Recent results from the first comprehensive molecular phylogeny of the *Ludwigia* genus (Liu et al. 2017) also provide clarification on clades of *Ludwigia* species that more recently originated in North America. These clades are all within the haplostemonous *Microcarpium* complex and are distantly related to the exotic invasive focal taxa (Liu et al. 2017).

*Ludwigia hexapetala* was first documented in the eastern and western United States in the mid-1800s and 1900s, respectively (Grewell et al. 2016a). The plant is naturalized in the southeastern and Pacific western states of the United States (Harms et al. 2017, WSNWCB. 2019), and Pennsylvania and New York (Grewell et al. 2016a). It is currently listed

as a *noxious weed* or *plant pest* for regulatory purposes by the states of California, Oregon, Washington, North Carolina, and South Carolina and is considered a plant pest in Florida and Virginia but without regulatory status. *Ludwigia hexapetala* is also naturalized in Australia, New Zealand, Europe, and Turkey (Thouvenot et al. 2013). To date, *L. grandiflora* (a single genotype of a hexaploid species) has a limited western distribution in coastal watersheds of southern California (Okada et al. 2009), although it is more widely distributed in southeastern United States and is highly invasive in Florida. The species has also been collected in Missouri (Zardini et al. 1991a). Erroneous reports of *L. grandiflora* in Oregon and Washington have been refuted with morphological, cytological, and molecular data (Grewell and Hoch, unpub. data). *Ludwigia grandiflora* is state-regulated as a *noxious weed* by South Carolina and Oregon. Although *L. grandiflora* has not been detected in Oregon, it is listed to prohibit sale and transport into the state, given the high potential risk of invasion. *Ludwigia grandiflora* is widely distributed in Europe and is considered a serious pest in many countries (EPPO 2011, Hussner 2012). In the United States, *L. peploides* subsp. *montevidensis* is naturalized in California, Oregon, Washington, Louisiana, and Oklahoma and in Europe, Cuba, Australia, and New Zealand; *L. peploides* subsp. *peploides* is found in the western and southern United States and is listed as *introduced* in eastern Australia and the Society Islands (Wagner and Hoch 2005, Hoch and Grewell 2012, Grewell et al. 2016a). *Ludwigia peploides* subsp. *glabrescens* is distributed in eastern and midwestern wetlands and, although now considered exotic, it is not invasive in parts of its naturalized range. However, in New York, *L. peploides* subsp. *glabrescens* is highly invasive in the Long Island Invasive Species Management Area. The Brooklyn Botanic Garden and Peconic Estuary Program provided data on invasiveness and recognized South American origin. The state of New York invasiveness rank for *L. peploides* subsp. *glabrescens* was designated as *very high* in 2008 because of its highly significant environmental impacts, high dispersal ability, and high difficulty to control (Jordan et al. 2008). *Ludwigia peploides* is state regulated as a *noxious weed* in Oregon and Washington (Butler 2019, WSNWCB 2019) and is listed as a *plant pest* in California. *Ludwigia decurrens* has often been considered a native species in southeastern United States where it is widely distributed. However, the five erect species of *Ludwigia* from sect. *Pterocaulon* T. P. Ramamoorthy & E. M. Zardini originate from Brazil, Argentina, Paraguay, and Uruguay and are naturalized in the southeast United States, Caribbean, Africa, Asia, and Europe (Wagner and Hoch 2005). *Ludwigia decurrens* is a state-regulated *noxious weed* in California. It was discovered in 2011 in a California rice field and has spread aggressively in irrigation canals, agricultural drains, rice fields, and natural wetlands (CDFA 2019).

## NATURE OF THE PROBLEM

### Ecological impacts

The primary ecological threat posed by invasive *Ludwigia* spp. is directly linked to high biomass production (Lambert

et al. 2010) that negatively affects resident vegetation and wildlife of invaded ecosystems. Once established, invasive *Ludwigia* spp. readily displace native vegetation through the formation of dense mats as well as growing over the surface of other floating aquatic plants (Grewell et al. 2016a). The total area invaded by invasive *Ludwigia* spp. in the United States is unknown, but rates of spread have been documented for *L. hexapetala* in the Laguna de Santa Rosa, in northern California, where the plant reached 100% cover over approximately 587 ha (1,450 acres) of the main channel in just 2 yr (Cal-IPC 2019). Most published studies on the ecological impacts of our focal taxa have come from Europe (Hussner 2009, 2010, EPPO 2011, Thouvenot et al. 2013), where the history of invasion stems from early 19th century introductions in southern France that have spread extensively (Dutartre 2004). However, recent results are confirming similar impacts within invaded aquatic ecosystems in the United States (e.g., Skaer Thomason et al. 2018a). Dense mats cause changes to ecological processes in a similar manner as other aquatic weeds, including reducing overall water flow patterns, accelerating sedimentation, slowing water circulation, lowering pH, and severely depleting dissolved oxygen concentrations in the water column (Dandelot et al. 2005a,b, Grewell et al. 2016a, Skaer Thomason et al. 2018a). Alteration of these physical and chemical aquatic characteristics have resulted in negative impacts on habitat quality, flora, and fauna (Dandelot et al. 2005b, Hussner 2009, Stiers et al. 2011). Dense mats outcompete and displace native plants, resulting in reduced species richness and abundance (Stiers et al. 2011). Additional evidence suggests that allelopathic effects of *Ludwigia* spp. may facilitate superior competitiveness over native plant species (Dandelot et al. 2008). Thiébaud et al. (2019) found that autoallelopathy by *L. hexapetala* could explain its invasiveness. In controlled experiments, results indicate allelochemical leachates from roots of *L. hexapetala* stimulated both shoot branching and synthesis of flavonols, which can facilitate establishment and may confer herbivore resistance (Thiébaud et al. 2019). *Ludwigia* spp. invasions can cause reductions in macroinvertebrate and fish populations (Stiers et al. 2011). Dense stands create a barrier for the movement of fish (Thouvenot et al. 2013) and degrade habitat quality for waterfowl and other migratory birds by displacing desirable wildlife, food plants, and open water habitat (Nehring and Kolthoff 2011, Grewell et al. 2016a).

### Economic impacts

*Ludwigia* spp. invasions also lead to social and economic problems. The formation of dense mats over and within the water column increase flood risk, constrain navigation, increase the cost of agricultural food production, and interfere with recreational activities, tourism, irrigation, and drainage (Thouvenot et al. 2013, Sarat et al. 2019). Economic data associated with *Ludwigia* spp. invasions are limited to the cost of controls. Traditional manual, mechanical, and chemical control methods are widely used to manage *Ludwigia* spp. but are costly. In the United States, the Division of Boating and Waterways spends \$7 million yr<sup>-1</sup> to control invasive plants, including *Ludwigia* spp., in



the Sacramento–San Joaquin River Delta in northern California (Brusati 2009). These control methods are also time consuming, physically difficult, and require continuous monitoring and repetitive treatments, adding to expense (Greenfield et al. 2006, Thouvenot et al. 2013, Hussner et al. 2016). Permits to apply herbicides for chemical control are also costly to obtain in the United States (Greenfield et al. 2006).

## MANAGEMENT APPROACHES

Management tools used by resource managers for suppressing *Ludwigia* spp. have included physical (manual uprooting/hand-pulling, tarping mowing, excavating, soil stripping/deep sediment removal, and mechanical harvesting), thermal (flaming), and chemical methods using aquatic herbicides. However, these options often provide short-term control requiring repeated annual treatments, have not been sustainable because of considerable on-going costs, and can be limited by regulatory restrictions (Sarat et al. 2015, Grewell et al. 2016a, Sarat et al. 2019). *Ludwigia hexapetala* and *L. peploides* produce viable seeds with a high capacity for germination under a wide range of temperatures (Gillard et al. 2017a,b), and they maintain persistent seedbanks that add to the need for long-term management pressure (Grewell et al. 2019b). Large field experiments were used to test mowing, tillage, and soil profile stripping to remove *Ludwigia* biomass and seed banks, followed by introduction of native species to resist reinvasion in the Isac Marshes (Loire-Atlantique region, France), but results were ineffective, with reinfestation of *L. hexapetala* to 90% of preresoration levels (Sarat et al. 2019). An alternative option to conventional control tactics includes classical biological control: the intentional introduction of host-specific natural enemies from an exotic pest's native range. Unlike conventional control methods, biological control has the potential to provide landscape-level suppression of weed populations and, if successful, is a long-term sustainable option (Van Driesche et al. 2010). Biological control is often a component of multifaceted integrated-management programs for invasive weeds because it often can be used alone or in combination with traditional approaches (Holtkamp 2004, Paynter and Flanagan 2004). The overall success rate of biological control programs, where complete control of the weed is reported, ranges from 25% (Moran et al. 2005) to 33% (McFadyen 2000). The rates are higher, 50 to 80%, when reported for individual countries or states (Fowler 2000, McFadyen 2000, Culliney 2005). However, even without complete control, biological control can provide suppression of plant spread and associated impacts leading to long-term economic and ecological effects (Wainger et al. 2018).

Along with uncertainty regarding the scale of impact of introduced agents, new classical biological control programs often have high initial costs (Fowler et al. 2000, McFadyen 2000, 2008). Program costs range from Australian \$0.6 to \$21.6 million for common ragweed (*Ambrosia artemisiifolia* L.) and catclaw mimosa (*Mimosa pigra* L.), respectively, in Australia (McFadyen 2008), and from United States \$100,000 to \$2.5 million for largeleaf lantana (*Lantana*

*camara* L.) and golden wattle (*Acacia pycnantha* Benth.), respectively, in South Africa (Van Wilgen et al. 2004). However, economic benefits from successful biological control programs can greatly exceed costs. Economic assessments of all biological control of weeds undertaken in Australia since 1903, including both successes and failures, demonstrated a benefit : cost ratio of 23 : 1 (McFadyen 2008). Returns on investment are more dramatic if benefit : cost ratios are estimated for individual or functional groups of weeds, which range from 34 : 1 to 4,333 : 1 (Van Wilgen 2004) and 50 : 1 to 3,726 : 1 (de Lange and Van Wilgen 2010), respectively.

Considering the uncertainty of success and the inherent opportunity costs, a thorough assessment should be conducted when selecting plants as targets for a biological control program. Existing literature and expert opinion should be considered regarding the weed's taxonomy, origins, habitat effects, and efficacy of existing control options, as described above (Harman 2004, Wheeler and Pemberton 2007, Paynter 2013). Information regarding the level of specificity required from biological control agents, known natural enemies of the weeds, areas surveyed, and potential conflicts of interest of a biological control program are also important considerations and will be discussed below.

## POTENTIAL RISKS AND RECOMMENDED SPECIES TO BE TESTED

The biogeographical relationships among *Ludwigia* species and taxa are complex. Polyploidy is extremely common in the genus, many species evolved through hybridization, and recent hybridization continues to be detected (Wagner et al. 2007, Liu et al. 2017, Grewell et al. unpub data). More extensive sampling and additional genetic studies are needed and are underway that will complement the evaluation of biological control.

The genus *Ludwigia* and related genera are well represented in North America. Clade A (Liu et al. 2017) is North American in origin, except for *ovalis* [*Ludwigia ovalis* Miq.] (East Asian). In Clade B, sect. *Jussiaea* is largely New World (South American) in origin, with the exception of some Old World endemics (Wagner et al. 2007, Liu et al. 2017). In the United States, there are 28 + described, native *Ludwigia* taxa (Liu et al. 2017), with most occurring in the southeastern United States, and 150 + native species across genera of the subfamily Onagroideae of the greater Onagraceae. For example, the *Ludwigia* sect. *Isnardia* (L.) W. L. Wagner & Hoch (Hoch et al. 2015) has five native species, including waterpurslane [*Ludwigia palustris* (L.) Elliott] and floating waterprimrose (*Ludwigia repens* J. R. Forst.), which overlap in range with our target exotic taxa, and *Ludwigia* sect. *Microcarpium* Peng includes 14 species, most exclusively North American in origin (Liu et al. 2017). To avoid unintended damage to nontarget species, we propose the initial development of a biological control should focus on the host range of a suitable agent restricted to sections *Jussiaea* and *Pterocaulon* in the genus *Ludwigia*.

In view of the numerous species in the genus *Ludwigia* and related genera and information from recent phyloge-

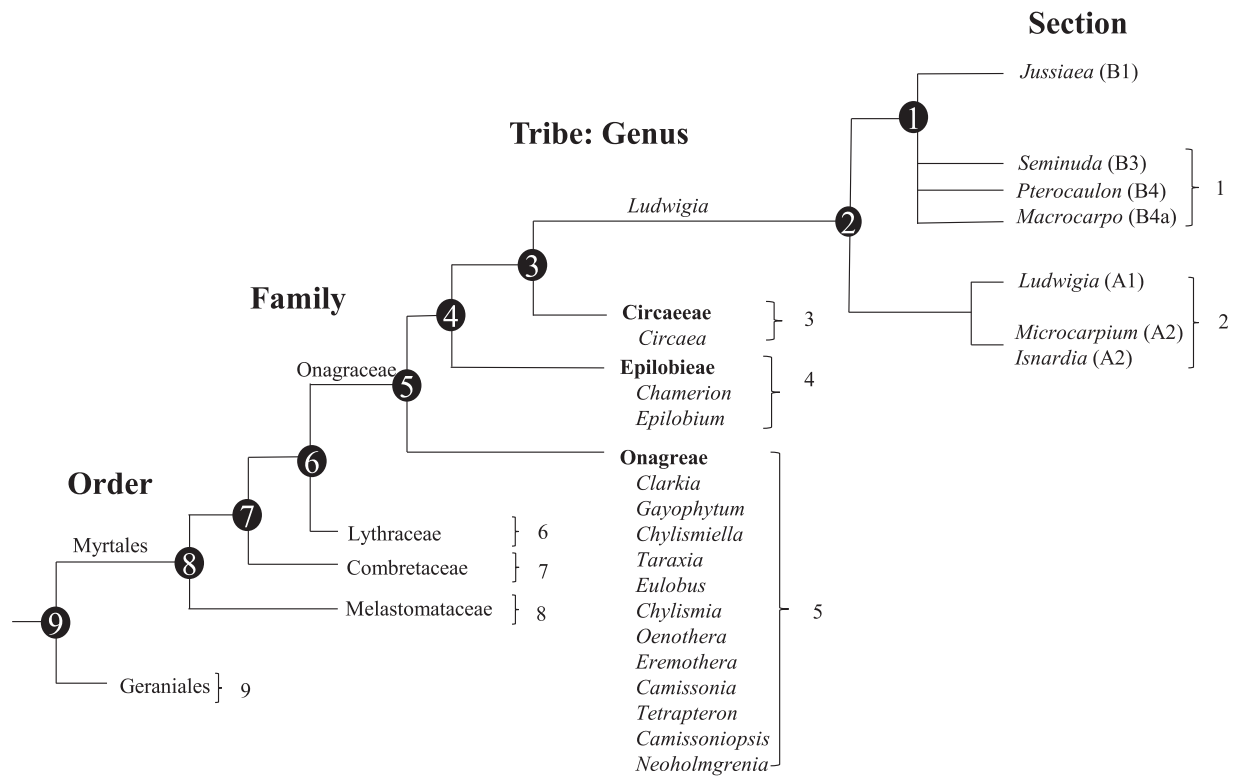


Figure 1. Phylogenetic relationship of representative Onagraceae (Levin et al. 2003, Wang et al. 2009, Liu et al 2017) used for host-specificity testing of candidate biological agents for the biological control of *Ludwigia* taxa. Numbered circles indicate nodes at which lineages diverge. Numbers on the right show the clade with increasing degrees of separation from the target weeds in sect. *Jussiaea* (*Ludwigia hexapetala*, *L. peploides* subsp. *peploides*, *L. peploides* subsp. *montevidensis*, and *L. grandiflora*)

netic (cladistic) analyses of the Onagraceae, we propose a plant test list that is based on relationships rather than nomenclature. The Wapshere (1974) centrifugal phylogenetic method (CPM) is the traditional method used to select plants for host-range tests. In that method, the candidate agent is exposed to a sequence of plant species from those most closely related to the target plant to those belonging to more-distant taxa. However, that method can be improved with the use of plant phylogenies instead of taxonomic circumscription (Briese 2003). Briese (2003) proposed a modernized version of CPM in which test plants are selected primarily based on their phylogenetic relationship to the target plant. We constructed our test list using the approach outlined by Briese (2003, 2005): plants were grouped into phylogenetic clades increasingly distant from the target plant, and relatedness to the target plant was determined as the “degrees of phylogenetic separation.” Separation occurs where a phylogenetic lineage branches into two distinct clades, and the higher the degree of separation, the fewer number of plant species tested from each clade (Figure 1). Because not all plants with strong taxonomic affinity to the target plant can be tested and to ensure that the plants selected maximize the measurement of risk posed by the candidate biological control agent, the representative species in each phylogenetic clade were those with biogeographical overlap and ecological similarity (i.e., life history, phenology, and growth) to the target plants. The following criteria were used to select plants for the test list

based on Briese and Walker (2008): 1) one to two representatives occurring in the United States from each of the two increasingly distant clades (i.e., section) within the genus *Ludwigia* for a total of 10 test species from the sections *Jussiaea*, *Seminuda*, *Pterocaulon*, *Macrocarpo*, *Ludwigia*, *Microcarpium*, and *Isnardia* (Table 1); 2) representatives from the related native genera (across three tribes) within the family Onagraceae (including threatened and endangered [T&E] species with protected status) occurring in the United States, and the number of test species per genus is based on genus size and T&E status for a total of 37 test plant species; 3) selected representatives of native genera belonging to the families Lythraceae, Combretaceae, and Melastomataceae (order Myrtales) occurring in the United States for a total of four plant species; and 4) one representative from a genus within the sister order Geraniales (Wang et al. 2009). The current test list of 52 test-plant species is the foundation for host-specificity testing of candidate agents targeting invasive *Ludwigia* spp. and is modifiable (i.e., to increase number of test plants species) as the biological control program evolves (Table 1).

In summary, the candidate target weeds belong to the sections *Jussiaea* and *Pterocaulon* in the genus *Ludwigia*. Suitable biological control agents must be host specific to the target weeds, but to be considered for introduction into the United States, they must not use any of the other native congeneric species of different taxonomic sections than the target taxa.

TABLE 1. PRELIMINARY LIST OF TEST PLANTS FOR HOST-SPECIFICITY TESTING OF BIOLOGICAL CONTROL AGENTS TARGETING EXOTIC *LUDWIGIA* SPP. IN THE UNITED STATES

Family	Degree of Separation	No. of Test Species	Test Species <sup>a</sup>
Onagraceae (Sect. <i>Jussiaea</i> )	0	targets	<i>Ludwigia hexapetala</i> , <i>L. grandiflora</i> , <i>L. peploides</i> subsp. <i>montevicensis</i> , <i>L. peploides</i> subsp. <i>peploides</i>
Onagraceae (Sect. <i>Seminuda</i> )	1	1	<i>L. leptocarpa</i>
Onagraceae (Sect. <i>Pterocaulon</i> )	1	1	<i>L. decurrens</i>
Onagraceae (Sect. <i>Macrocarpo</i> )	1	2	<i>L. octovalvis</i> and <i>L. bonariensis</i>
Onagraceae (Sect. <i>Ludwigia</i> )	2	2	<i>L. alternifolia</i> and <i>L. hirtella</i>
Onagraceae (Sect. <i>Microcarpum</i> )	2	2	<i>L. glandulosa</i> and <i>L. polycarpa</i>
Onagraceae (Sect. <i>Isardia</i> )	2	2	<i>L. repens</i> and <i>L. palustris</i>
Onagraceae (Tribe Circaeae)	3	2	<i>Circaea alpina</i> subsp. <i>pacifica</i> and <i>C. canadensis</i>
Onagraceae (Tribe Epilobieae)	4	2	<i>Chamerion angustifolium</i> subsp. <i>circumvagum</i> and <i>C. latifolium</i>
	4	3	<i>Epilobium ciliatum</i> subsp. <i>ciliatum</i> , <i>E. canum</i> , <i>E. brachycarpum</i>
Onagraceae (Tribe Onagreae)	5	6	<i>Clarkia amoena</i> and <i>C. unguiculata</i> . T&E species: <i>C. franciscana</i> , <i>C. imbricata</i> , <i>C. speciosa</i> subsp. <i>immaculata</i> , and <i>C. springvillensis</i>
	5	2	<i>Gayophytum humile</i> and <i>G. racemosum</i>
	5	1	<i>Chylismiella pterosperma</i>
	5	2	<i>Taraxia subacaulis</i> and <i>T. tanacetifolia</i>
	5	1	<i>Eulobus californicus</i>
	5	2	<i>Chylismia brevipes</i> subsp. <i>brevipes</i> and <i>C. claviformis</i> <sup>b</sup>
	5	5	<i>Oenothera elata</i> subsp. <i>hookeri</i> , <i>O. curtiflora</i> , and T&E species: <i>O. coloradensis</i> subsp. <i>coloradensis</i> , <i>O. californica</i> subsp. <i>eurekaensis</i> , and <i>O. deltoides</i> subsp. <i>howellii</i>
	5	2	<i>Eremothera refracta</i> and <i>E. boothii</i> subsp. <i>decorticans</i> <sup>b</sup>
	5	3	<i>Camissonia campestris</i> subsp. <i>campestris</i> , <i>C. contorta</i> , <i>C. benitensis</i> (T&E) <sup>b</sup>
	5	2	<i>Tetrapterum graciliflorum</i> and <i>T. palmeri</i>
	5	2	<i>Camissoniopsis hirtella</i> and <i>C. ignota</i>
	5	2	<i>Neoholmgrenia andina</i> and <i>N. hilgardii</i>
Lythraceae	6	2	<i>Ammannia coccinea</i> , <i>Lythrum californicum</i> , or <i>Rotala ramosior</i>
Combretaceae	7	1	<i>Conocarpus erectus</i> or <i>Laguncularia racemosa</i>
Melastomataceae	8	1	<i>Rhexia cubensis</i> , <i>R. petiolata</i> , or <i>Tetrazygia bicolor</i>
Geraniaceae (Order Geraniales)	9	1	<i>Geranium carolinianum</i>

Abbreviation: T&E, threatened and endangered.

<sup>a</sup>Genus occurs in dry habitats.

<sup>b</sup>Test plants listed by decreasing phylogenetic relatedness (degrees of separation) to the target weeds in sect. *Jussiaea*.

## NATURAL ENEMIES IN THE INTRODUCED RANGE

Surveys have been conducted in the United States to document the identity of indigenous herbivores, arthropods, and pathogens, associated with *Ludwigia* spp. (Harms and Grodowitz 2009, Harms and Grodowitz 2012, Harms et al. 2017). In the southeastern United States, three species of leaf beetles (Coleoptera: Chrysomelidae: *Lysathia ludoviciana* Fall, *Altica litigata* Fall, and a *Chaetocnema* Stephens sp.), four species of weevils (Coleoptera: Curculionidae: *Tyloderma sphaerocarpha* Wibmer, *Perigaster cretura* Herbst, and two *Auleutes* Dietz spp.), and two species of caterpillars (*Eumorphia* Hübner sp. [Lepidoptera: Sphingidae] and *Spilosoma* Curtis [Lepidoptera: Arctiidae]) feed on *L. peploides* (Clark 1976, Campbell and Clark 1983, Haag et al. 1986, Harms and Grodowitz 2012). Voracious but transitory herbivory by larvae and adults of the flea beetle *L. ludoviciana* and *A. litigata* has been documented on *L. grandiflora* in Alabama (McGregor et al. 1996) and on *L. hexapetala* in California (Carruthers et al. 2011), respectively. In Alabama, 14 insect herbivore species, including *P. cretura*, and 7 fungal taxa were associated with *L. hexapetala* (Harms et al. 2017). However, despite the diverse assemblage of herbivores on *L. hexapetala*, damage was relatively low. Furthermore, most of the herbivore species collected were generalist and known to feed on other *Ludwigia* spp.

## AREAS SURVEYED FOR NATURAL ENEMIES

The first foreign exploration for natural enemies of *Ludwigia* spp. was conducted in Argentina, Paraguay, and Brazil by Cordo and DeLoach (1982a,b). Those surveys were conducted in the northeastern region of Argentina from the Buenos Aires province to the Formosa province bordering Paraguay; Asunción, Paraguay near the north border of Argentina; and Barão de Melgaço, Mato Grosso, in the central west region of Brazil (Figure 2). A recent in-depth survey conducted by scientists from the Fundación para el Estudio de Especies Invasivas (FuEDEI) covered north central and eastern Argentina, where the main subtropical and temperate wetlands are located (Hernández and Cabrera Walsh 2014). Insects were sampled at 41 sites, sometimes more than once, from spring to fall over a 3-yr period. The Buenos Aires and Entre Ríos provinces in Argentina were surveyed once more by FuEDEI and the US Department of Agriculture (USDA) scientists in the late summer of 2019, followed by the first surveys conducted in coastal and central locations in Uruguay (Figure 2). The surveys in Uruguay were conducted in collaboration with scientists from the Instituto Nacional de Investigación Agropecuaria (INIA).

## NATURAL ENEMIES FOUND

Cordo and DeLoach (1982a,b) collected one leaf beetle and four weevil species from *L. peploides* in Argentina:

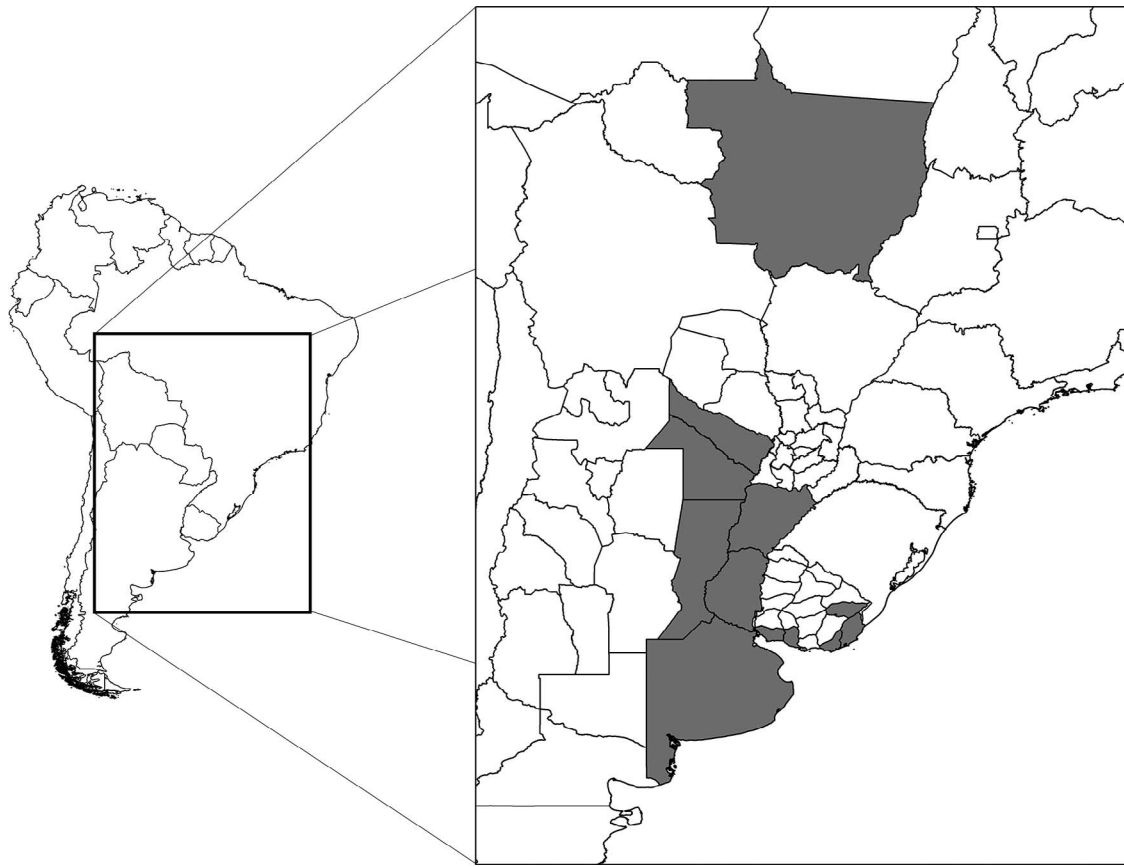


Figure 2. Geographic range (shaded) of foreign exploration of natural enemies of *Ludwigia* taxa in South America.

*Lysathia flavipes* (Boheman) (Coleoptera: Chrysomelidae), two *Tyloclerma* spp. (Coleoptera: Curculionidae), *Sudauleutes bosqi* Hustache (Coleoptera: Curculionidae; formerly *Auleutes bosqi* Hustache [Colonnelli 2004]), and an *Onychylis* LeConte sp. nr. *nigrirostris* (Boheman) (Coleoptera: Curculionidae). The adult stage of all species feed on leaves. The larvae of the *Tyloclerma* spp. and *O. nigrirostris* are stem borers, and the larvae of *S. bosqi* feed on developing leaves at the meristem.

Hernández and Cabrera Walsh (2014) collected insect herbivores from *L. peploides* subsp. *montevideensis*, *L. hexapetala*, and *L. grandiflora*. They found 19 insect species, across six feeding guilds, feeding on *L. hexapetala*. Of those species, only two species were also found on *L. grandiflora* and one on *L. peploides*. The list of promising biological control agents includes a thrips species (*Liothrips ludwigi* Zamar [Thysanoptera: Phlaeothripidae]), six stem-boring beetle species (*Meroenemus binotatus* Boheman) and five *Tyloclerma* spp. [Coleoptera: Curculionidae], and one fruit-feeding weevil (*Tyloclerma nigromaculatum* Hustache [Coleoptera: Curculionidae]). Of the five *Tyloclerma* spp., three have been identified to species: *Tyloclerma affine* Wibmer, *Tyloclerma longisquamum* Wibmer, and *Tyloclerma elongate* Wibmer (Hernández and Cabrera Walsh 2014). *Liothrips ludwigi* is a cell-content feeder that feeds and breeds in the apical buds of the plants. Adults and larvae share the same refuge-feeding sites in young leaves. The thrips species also feeds on *L. grandiflora* and *L. peploides* subsp. *montevideensis* (Hernández and Cabrera Walsh 2014, FuEDEI 2015, FuEDEI 2017). Hernández and Cabrera

Walsh (2014) also found *Galerucella obliterata* Oliver (Coleoptera: Chrysomelidae) on *L. grandiflora* and *S. bosqi* on *L. grandiflora* and *L. peploides*, but they were not included among the most-promising candidate biological control agents because they were not sufficiently host specific.

Prioritization of natural enemies for further research may be influenced by the type of feeding damage inflicted on the target plant. Recent evidence indicates that, although *L. hexapetala* and *L. grandiflora* are capable of sexual reproduction (Gillard et al. 2017a) and asexual (clonal) reproduction via water dispersal of vegetative fragments (Skaer Thomason et al. 2018b), the invasive spread of both species in the United States is predominately clonal (Okada et al. 2009). Seasonal fluctuations in reproduction and growth rates among candidate target weeds result in high biomass production throughout the growing season, resulting in rapid weed coverage and associated ecological and economic damage (Grewell et al. 2016b). Those findings suggest that focusing biological control herbivory on plant components that contribute to spread and vegetative growth may reduce the negative consequences of *Ludwigia* invasions. Both stem boring and defoliating insect species have been identified as potential biological control agents previously and may contribute to the reduction of plant biomass production (Hernández and Cabrera Walsh 2014). However, the biology and host specificity of these insects to native *Ludwigia* species in the United States is unknown and needs to be established.



## POTENTIAL CONFLICTS OF INTEREST

Conflicts of interest in weed biological control arise when the target weed is associated with economic and/or ecological benefits and with nontarget effects of biological control. However, the risk of conflicts that may delay initiation of the biological control program against *Ludwigia* spp. is low. The target *Ludwigia* taxa have no value to the horticultural industry given their status as noxious weeds in several states where the sale, propagation, and distribution is prohibited. Likewise, the target taxa have questionable ecological value, although honey bees may acquire some nutritional benefit from their summer blooms. The many negative ecological impacts attributed to the target taxa (as previously described) and the low damage by native insect herbivores indicates that the weeds are not an important source of food and habitat for native wildlife. Furthermore, nearly all *Ludwigia* species in sect. *Jussiaea* can form interspecific hybrids, which have been found in the native range (Zardini et al. 1991b) and in the United States (Grewell and Gaskin, unpub. data). Thus, additional research is needed to understand hybridization among exotic and native species, and host-specificity testing trials should include known hybrids to determine the relevance of hybridization to a herbivore's host range. Nontarget conflicts may occur because two close relatives of the target taxa (*L. repens* and *L. palustris*) are native to the United States, are habitat associates with the target weeds, and are sold (advertised as *L. repens* and *L. palustris*) as ornamentals for freshwater aquariums. The risk of nontarget feeding on these species, however, must be quantified for candidate agents during host-specificity testing to avoid unintended damage to economically important or native plant species.

## RECOMMENDATIONS AND CONCLUSIONS

Biological control programs targeting aquatic and riparian weeds are less common than terrestrial weeds, but several have resulted in success. For example, alligatorweed [*Alternanthera philoxeroides* (Mart.) Griseb.] and waterhyacinth [*Pontederia crassipes* Mart. (Pellegrini et al. 2018), formerly *Eichhornia crassipes* [Mart.] Solms) have been effectively controlled or reduced in certain regions of the southern United States (Buckingham 2002, Center et al. 2002) and other warm regions of the world where the plants were a problem (Wilson et al. 2007, Van Driesche et al. 2010). The niche breadth, multiple growth forms, and ecology of *L. hexapetala* is very similar to that of alligatorweed, although they are unrelated, which suggests biological control may be feasible or not limited by aquatic environmental factors. Overall, the average effect of programs targeting aquatic or wetland weeds is greater than that of programs targeting terrestrial weeds. Paynter et al. (2012) predicted biological control impact using evidence-based criteria and quantitative data. Based on that predictive framework, invasive *Ludwigia* taxa are "good target" weeds for biological control because they possess attractive combinations of three factors: they are aquatic, populations are clonal, and they are not major weeds in their natural habitats in the native ranges. Furthermore, genotypic diversity of the polyploid species is very low in the United

States, which holds promise for broad interpretations of results from host testing. For example, multiple invasive populations of *L. hexapetala* in Alabama, California, Oregon, Washington, and Florida are a single genotype (Grewell and Gaskin, unpub. data).

Additional factors that lend support to targeting *Ludwigia* taxa for biological control include a broad foundational knowledge base on which to develop the program. Numerous surveys for herbivores have been conducted and various natural enemies identified, including species that partition their attack among leaves, flowers, and stems. Facets of these herbivore's biology and known hosts have been compiled where available, which facilitates prioritization of candidates to be considered (Cordo and DeLoach 1982a,b, Hernández and Cabrera Walsh 2014). Teams of scientists from the weed's native and exotic ranges are already collaborating, which can expedite the research. A growing body of literature on the invasion and biology of select species in the United States, Europe, and the native range also provides critical baseline data on plant population dynamics and distributions among regions (Okada et al. 2009, Hussner 2010, Hussner 2012, Thouvenot et al. 2013, Grewell et al. 2016a,b., Gillard et al. 2017b, Skaer Thomason et al. 2018a,b, Grewell et al. 2019a,b, Thiébaud et al. 2019).

Stakeholder support for the development of a biological control program remains strong because this may be the only landscape-level control option available for some resource managers. Naturalized, invasive *Ludwigia* spp. are widespread throughout the Pacific western and southeastern states, and several states have categorized one or more of the candidate target taxa as *noxious weeds* or *plant pests*. For those weeds in which eradication is no longer possible because of regional abundance, but control (i.e., prevention of dispersal) is mandated by law, biological control may be the primary control method when management by traditional methods is not feasible in ecologically sensitive systems.

Although these factors suggest that invasive *Ludwigia* spp. are suitable targets for weed biological control, interspecific hybridization needs further investigation for a comprehensive evaluation of potential natural enemies. Although hybridization between an exotic and native species is unlikely due to phylogenetic distances, if native by exotic hybrids are possible and detected, it could complicate the likelihood of discovering a natural enemy that is sufficiently host specific to discriminate between exotics and hybrids. To date, all recent hybrids have been formed from two exotic *Ludwigia* species from the same section of the genus. Additionally, although updated phylogenetic analyses (Liu et al. 2017) have greatly increased knowledge and support for this project, unresolved uncertainty of the distribution and genetic variation of invasive *Ludwigia* spp. requires additional research to adequately interpret host-range results.

Despite these limitations, stakeholders continue to request biological control tools for *Ludwigia* invasions, and the authors conclude that the development of a biological control program for invasive *Ludwigia* spp. is warranted based on the potential benefits of a successful program. The



initial effort will focus on invaders from sect. *Jussiaea* and will, hopefully, expand to consider management tools for exotic *L. decurrens*. The opportunity costs associated with developing a program that fails to provide adequate control is not trivial but is balanced by the existing knowledge of the system combined with the magnitude of the growing problem. The authors, therefore, propose to proceed cautiously by expanding research to understand the biology and evolution of the target weeds. Additionally, focus will be placed on understanding the host ranges of high-priority insects based on published literature and expert opinion (Cordo and DeLoach 1982a,b, Hernández and Cabrera Walsh 2014, FuEDEI 2015, FuEDEI 2017). Initial testing will begin with *L. ludwigi*, a thrips species for which the Argentina government has already provided an export permit. Similarly, the Uruguayan government has provided an export permit for *Tyloderma* species, and assessment will also begin with these species. It is expected that new herbivores will be discovered in future surveys of the target weed's native ranges, and these will also be considered. Thus far, the efforts to control invasive *Ludwigia* spp. using traditional methods have failed to provide long-term, landscape-level suppression, so new management approaches are necessary.

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## LITERATURE CITED

Briese DT. 2003. The centrifugal phylogenetic method used to select plants for host-specificity testing of weed biological control agents: Can and should it be modified? pp. 23–33. In: H. Spafford Jacob, D.T. Briese (eds.). Improving the selection, testing and evaluation of weed biological control agents. CRC for Australia Weed Management, Technical Series No. 7., Indooroopilly, QLD, Australia.

Briese DT. 2005. Translating host-specificity test results into the real world: The need to harmonize the yin and yang of current testing procedures. *Biol. Control* 35:208–214.

Briese DT, Walker A. 2008. Choosing the right plants to test: The host-specificity of *Longitarsus* sp. (Coleoptera: Chrysomelidae) a potential biological control agent of *Heliotropium amplexicaule*. *Biol. Control* 44:271–285.

Brusati E. 2009. The cost of weeds to California. *Calif Invasive Plant Council. Newsl* 17:6–7.

Buckingham GR. 2002. Alligatorweed, pp. 5–15. In: R. Van Driesche (ed.). Biological control of invasive plants in the eastern United States. USDA Forest Service Publication FHTET-2002-04, Morgantown, WV.

Butler T. 2019. Noxious weed policy and classification system. Noxious Weed Program, Oregon Department of Agriculture, Salem, OR. 13 pp. [Cal-IPC] California Invasive Plant Council 2019. California Invasive Plant Inventory. California Invasive Plant Council, Berkeley, CA. <https://www.cal-ipc.org/plants/inventory/>. Accessed April 14, 2019.

Campbell JM, Clark WJ. 1983. Observations on host selection by *Lysathia ludoviciana* (Chrysomelidae), a beetle with potential for biological control of certain aquatic weeds. *Tex. J. Sci.* 35:165–167.

Carruthers I, Franc M, Gee WS, Cossé AA, Grewell BJ, Beck J. 2011. Volatile emissions from the flea beetle *Altica litigata* (Coleoptera: Chrysomelidae) associated with invasive *Ludwigia hexapetala*. *Chemoecology* 21:253–259.

[CDFA] California Department of Agriculture. 2019. Noxious Weeds List. [http://www.cdffa.ca.gov/plant/IPC/encyclopedia/weedinfo/winfo\\_table-sciname.html#](http://www.cdffa.ca.gov/plant/IPC/encyclopedia/weedinfo/winfo_table-sciname.html#). Accessed October 10, 2019.

Center TD, Hill MP, Cordo H, Julien MH. 2002. Waterhyacinth, pp. 41–63. In: R. Van Driesche, B. Blossey, M. Hoddle, S. Lyon, R. Reardon (eds.). Biological control of invasive plants in the eastern United States. USDA Forest Service Publication FHTET-2002-04, Morgantown, WV.

Clark WE. 1976. Notes on the life history and habitats of *Perigaster cretura* (Herbst) (Coleoptera: Curculionidae) with descriptions of the larva and pupa. *Coleopt. Bull.* 30:159–165.

Colonnelli E. 2004. Catalogue of Ceutorhynchinae of the world, with a key to genera (Insecta: Coleoptera: Curculionidae). Argania Editio, S.C.P., Barcelona, Spain. 124 pp.

Cordo HA, DeLoach CJ. 1982a. Notes on the weevils *Tyloderma*, *Auletes*, and other aquatic plants in the southern South America. *Coleopt. Bull.* 36:291–297.

Cordo HA, DeLoach CJ. 1982b. The flea beetle, *Lysathia flavipes*, that attacks *Ludwigia* (water primrose) and *Myriophyllum* (Parrotfeather) in Argentina. *Coleopt. Bull.* 36:299–302.

Cronk QC, Fuller JL. 1995. Plant invaders: The threat to natural ecosystems. Chapman and Hall, London. 241 pp.

Culliney TW. 2005. Benefits of classical biological control for managing invasive plants. *Crit. Rev. Plant Sci.* 24:131–150.

Dandelot S, Matheron R, Le Petit J, Verlaque R, Cazaubon A. 2005a. Temporal variations of physicochemical and microbiological parameters in three freshwater ecosystems (southeastern France) invaded by *Ludwigia* spp. *C. R. Biol.* 328:991–999. [in French]

Dandelot S, Robles C, Pech N, Cazaubon A, Verlaque R. 2008. Allelopathic potential of two invasive *Ludwigia* spp. *Aquat. Bot.* 88:311–316.

Dandelot S, Verlaque R, Dutartre A, Cazaubon A. 2005b. Ecological, dynamic and taxonomic problems due to *Ludwigia* (Onagraceae) in France. *Hydrobiologia* 551:131–136.

de Lange WJ, van Wilgen BW. 2010. An economic assessment of the contribution of biological control to the management of invasive alien plants and to the protection of ecosystem services in South Africa. *Biol. Invasions* 12:4113–4124.

Dutartre A. 2004. “*Ludwigia peploides* (Kunth.) P.H. Raven, *Ludwigia grandiflora* (Mxhaux) Greuter & Burdet. Les jussies,” pp. 76–81. In: S. Muller (ed.) *Plantes invasives en France (Patrimoines naturels, 62)*. Museum National d'Histoire Naturelle, Paris. [in French]

[EPP] European and Mediterranean Plant Protection Organization. 2011. *Ludwigia grandiflora* and *Ludwigia peploides* Onagraceae—Water primroses. *Eur. Mediterr. Plant Prot. Organ. Bull.* 41:414–418.

Fowler SV. 2000. Trivial and political reasons for the failure of classical biological control of weeds: a personal view, pp. 169–172. In: NR Spencer (ed.). Proceedings of the X International Symposium on Biological Control of Weeds. Advanced Litho Printing, Great Falls, MT.

Fowler SV, Syrett P, Hill R. 2000. Success and safety in the biological control of environmental weeds in New Zealand. *Austral Ecol.* 25:553–562.

[FuEDEI] Fundación para el Estudio de Especies Invasivas. 2015. Annual report 2015. FuEDEI, Hurlingham, Argentina. 51 pp.

[FuEDEI] Fundación para el Estudio de Especies Invasivas. 2017. Annual report 2015–17. FuEDEI, Hurlingham, Argentina. 118 pp.

Gallardo B, Clavero M, Sánchez MI, Vilà M. 2016. Global ecological impacts of invasive species in aquatic ecosystems. *Glob. Change Biol.* 22:151–163.

Gillard M, Grewell BJ, Futrell CJ, Deleu C, Thiébaud G. 2017a. Germination and seedling growth of water primroses: A cross experiment between two invaded ranges with contrasting climates. *Front. Plant. Sci.* 8:1677.

Gillard M, Thiébaud G, Deleu C, Leroy D. 2017b. Present and future distribution of three aquatic plants taxa across the world: decrease in native and increases in invasive ranges. *Biol. Invasions* 19:2159–2170.

Greenfield BK, Blankinship M, McNabb TJ. 2006. Control costs, operation, and permitting issues for non-chemical plant control: Case studies in the San Francisco Bay-Delta region, California. *J. Aquat. Plant Manag.* 44:40–49.

Grewell BJ, Futrell CJ, Iannucci MT, Drenovksy RE. 2019a. Resprouting potential of rhizome fragments from invasive macrophyte reveals

- superior colonization ability of the diploid congener. *AoB Plants* 11:plz071. doi: 10.1093/aobpla/plz071.
- Grewell BJ, Gillard MB, Futrell CJ, Castillo JM. 2019b. Seedling emergence from seed banks in *Ludwigia hexapetala*-invaded wetlands: implications for restoration. *Plants*, 8:451. doi:10.3390/plants8110451. [Special issue Seed Behavior in Soil]
- Grewell BJ, Netherland MD, Skaer Thomason MJS. 2016a. Establishing research and management priorities for invasive water primroses (*Ludwigia* spp.). ERDC/EL TR-162 Technical Report. U.S. Army Engineer Research and Development Center, Washington DC. 44 pp.
- Grewell BJ, Skaer Thomason MJ, Futrell CJ, Iannucci M, Drenovsky RE. 2016b. Trait responses of native aquatic macrophyte congeners: colonizing diploid outperforms polyploid. *AoB Plants* 8:plw014. doi.org/10.1093/aobpla/plw014.
- Haag KH, Habeck DH, Buckingham GR. 1986. Native insect enemies of aquatic macrophytes other than moths and beetles. *Aquatics* 8:16–22.
- Harman HM. 2004. Feasibility of biological control of grey willow *Salix cinerea*. Department of Conservation, DOC Science Internal Series 183, Wellington, New Zealand. 29 pp.
- Harms NE, Grodowitz MJ. 2009. Insect herbivores of aquatic and wetland plants in the United States: A checklist from literature. *J. Aquat. Plant Manag.* 47:73–96.
- Harms NE, Grodowitz MJ. 2012. Herbivorous insects associated with *Ludwigia peploides* (Onagraceae) in the southern United States. *Southwest. Nat.* 57:123–127.
- Harms NE, Shearer JF, Grodowitz MJ. 2017. Folivory and disease occurrence on *Ludwigia hexapetala* in Guntersville reservoir, Alabama. *J. Aquat. Plant Manag.* 55:19–25.
- Hernández MC, Cabrera Walsh G. 2014. Insect herbivores associated with *Ludwigia* species, *Oligospermum* section, in their Argentine distribution. *J. Insect Sci* (Tucson). 14(201).
- Hickman JC. (ed.) 1993. *The Jepson manual: Higher plants of California*. University of California Press, Berkeley, CA. 1400 pp.
- Hoch PC, Grewell BJ. 2012. *Ludwigia*. In: *Jepson Flora Project* (eds.) *Jepson eFlora*. [http://ucjeps.berkeley.edu/eflora/eflora\\_display.php?tid=9765](http://ucjeps.berkeley.edu/eflora/eflora_display.php?tid=9765). Accessed March 14, 2018.
- Hoch PC, Wagner LW, Raven P. 2015. The correct name for section *Ludwigia* L. (Onagraceae). *Phytokeys* 50:31–34. [Short communication]
- Holtkamp RH. 2004. The role of biological control agents in an IWM program for *Chrysanthemoides monilifera* subsp. *rotundata* (bitou bush), pp. 412–414. In: J. M. Cullen, D. T. Briese, D. J. Kriticos, W. M. Lonsdale, L. Morin, J. K. Scott (eds.). *Proceedings of the XI International Symposium on Biological Control of Weeds*. CSIRO Entomology, Canberra, Australia.
- Hussner A. 2009. Growth and photosynthesis of four invasive aquatic plant species in Europe. *Weed Res.* 49:506–515.
- Hussner A. 2010. Growth response and root system development of the invasive *Ludwigia grandiflora* and *Ludwigia peploides* to nutrient availability and water level. *Fundam. Appl. Limnol. Arch. Hydrobiol.* 177:189–196.
- Hussner A. 2012. Alien aquatic plant species in European countries. *Weed Res.* 52:297–306.
- Hussner A, Windhaust M, Starfinger U. 2016. From weed biology to successful control: an example of successful management of *Ludwigia grandiflora* in Germany. *Eur. Weed Res.* 56:434–441.
- Jordan MJ, Moore G, Weldy TW. 2008. Invasiveness ranking system for non-native plants of New York. *Ludwigia peploides* (Kunth) Raven ssp. *glabrescens* (Kuntze) Raven. Unpublished. Non-Native Plant Invasiveness Ranking Form. The Nature Conservancy, Cold Spring Harbor, NY; Brooklyn Botanic Garden, Brooklyn, NY; The Nature Conservancy, Albany, NY. 11 pp.
- Lambert E, Dutartre A, Coudreuse J. 2010. Relationships between the biomass production of invasive *Ludwigia* species and physical properties of habitats in France. *Hydrobiologia* 656:173–186.
- Levin RA, Wagner WL, Hoch PC, Nepokroeff M, Pires JC, Zimmer EA, Systsma KJ. 2003. Family-level relationships of Onagraceae based on chloroplast *RBCL* and *NDHF* data. *Am. J. Bot.* 90:107–115.
- Liu S, Hoch P, Diazgranados M, Raven PH, Barber JC. 2017. Multi-locus phylogeny of *Ludwigia* (Onagraceae): Insights on infra-generic relationships and the current classification of the genus. *Taxon* 66:1112–1127.
- Lodge DM, Stein RA, Brown KM, Govich AP, Brönmark C, Garvey JE, Klosiewski SP. 1998. Predicting impact of freshwater exotic species on native biodiversity: Challenges in spatial scaling. *Aust. J. Ecol.* 23:53–67.
- McFadyen RE. 2000. Success in biological control of weeds, pp. 3–14. In: N. R. Spencer (ed.). *Proceedings of the X International Symposium on Biological Control of Weeds*. Advanced Litho Printing, Great Falls, MT.
- McFadyen RE. 2008. Return on investment: determining economic impact of biological control programmes, pp. 67–74. In: M. H. Julien (eds.). *Proceedings of the XII International Symposium on Biological Control of Weeds*. CAB, Wallingford, UK.
- McGregor MA, Bayne DR, Steeger JG, Webber EC, Reutebuch E. 1996. The potential for biological control of water primrose (*Ludwigia grandiflora*) by the water primrose flea beetle (*Lysathia ludoviciana*) in the southeastern United States. *J. Aquat. Plant Manag.* 34:74–76.
- Moran VC, Hoffmann JH, Zimmermann HG. 2005. Biological control of invasive alien plants in South Africa: necessary, circumspection, and success. *Front. Ecol. Environ.* 3:71–77.
- Munz PA. 1942. *Studies in Onagraceae XII: A revision of the new world species of Jussiaea*. *Darwiniana* (San Isidro) 4:179–284.
- Nehring S, Kolthoff D. 2011. The invasive water primrose *Ludwigia grandiflora* (Michaux) Greuter & Burdet (Spermatophyta: Onagraceae) in Germany: First record and ecological risk assessment. *Aquat. Invasions* 6:83–89.
- Nesom GL, Kartesz JT. 2000. Observations on the *Ludwigia uruguayensis* complex (Onagraceae) in the United States. *Castanea* 65:123–125.
- Okada M, Grewell BJ, Jasieniuk M. 2009. Clonal spread of invasive *Ludwigia hexapetala* and *Ludwigia grandiflora* in freshwater wetlands of California. *Aquat. Bot.* 91:123–129.
- Paynter Q. 2013. Feasibility of biocontrol of *Lagarosiphon major* in New Zealand. *Landcare Research, Manaaki Whenua, New Zealand*. 17 pp.
- Paynter Q, Flanagan GJ. 2004. Integrated weed management—Could we be doing better? Lessons from controlling the invasive wetland shrub, *Mimosa pigra*, pp. 361–369. In: J. M. Cullen, D. T. Briese, D. J. Kriticos, W. M. Lonsdale, L. Morin, J. K. Scott (eds.). *Proceedings of the XI International Symposium on Biological Control of Weeds*. CSIRO Entomology, Canberra, Australia.
- Paynter Q, Overton JC, Hill RL, Bellgard SE, Dawson MI. 2012. Plant traits predict the success of weed biocontrol. *J. Appl. Ecol.* 49:1140–1148.
- Pellegrini MOO, Horn CN, Almeida RE. 2018. Total evidence phylogeny of Pontederiaceae (Commelinales) sheds light on the necessity of its recircumscription and synopsis of *Pontederia* L. *Phytokeys* 108:25–83.
- Pesamosca SC, Boldrini IL. 2015. *Ludwigia litoranea* (Onagraceae), a new species from coastal southern Brazil. *Phytotaxa* 230:183–188.
- Raven PH. 1963a. The old world species of *Ludwigia* (including *Jussiaea*), with a synopsis of the genus (Onagraceae). *Reinwardtia* 6:327–427.
- Raven PH. 1963b. Amphitropical relationships in the floras of north and south America. *Q. Rev. Biol.* 38:151–177.
- Raven PH, Tai W. 1979. Observations of chromosomes in *Ludwigia* (Onagraceae). *Ann. Mo. Bot. Gard.* 66:862–879.
- Reichard SH, White P. 2000. Horticulture as a pathway of invasive plant introductions in the United States. *Bioscience* 51:103–113.
- Sarat E, Blottière D, Dutartre A, Poulet N, Soubeyran Y. 2019. Water primrose (*Ludwigia* spp.), pp. 65–88. In: E. Sarat, D. Blottière, A. Dutartre, N. Poulet, Y. Soubeyran (eds.). *Invasive alien species in aquatic environments: Practical information and management insights*. Volume 3. *Management Insights (cont.)*. French Biodiversity Agency (AFB) Knowledge for Action Series and French Committee for International Union for Conservation of Nature (IUCN), Paris.
- Sarat E, Mazaubert E, Dutartre A, Poulet N, Soubeyran Y. 2015. Water primrose (*Ludwigia* spp.), pp. 55–73. In: E. Sarat, E. Mazaubert, A. Dutartre, N. Poulet, Y. Soubeyran Y (eds.). *Invasive alien species in aquatic environments: Practical information and management insights*. Vol 2. *Management Insights*. ONEMA (French National Agency for Water and Aquatic Environments), Knowledge for Action Series, Paris.
- Skaer Thomason MJ, Grewell BJ, Netherland MD. 2018a. Dynamics of *Ludwigia hexapetala* invasion at three spatial scales in a regulated river. *Wetlands* 38:1285–1298.
- Skaer Thomason MJ, McCort CD, Netherland MD, Grewell B. 2018b. Temporal and nonlinear dispersal patterns of *Ludwigia hexapetala* in a regulated river. *Wetlands Ecol. Manag.* 26:751–762.
- Stiers I, Crohain N, Josens G, Triest L. 2011. Impact of three aquatic invasive species on native plants and macroinvertebrates in temperate ponds. *Biol. Invasions* 13:2715–2726.
- Thiébaud G., Tarayre M, Rodríguez-Pérez H. 2019. Allelopathic effects of native versus invasive plants on one major invader. *Front. Plant Sci.* 10:854.

- Thouvenot L, Haury J, Thiébaud G. 2013. A success story: Water primroses, aquatic plant pests. *Aquat. Conserv. Mar. Freshw. Ecosyst.* 23:790–803.
- van Driesche RG, Carruthers RI, Center T, Hoddle MS, Hough-Goldstein J, Morin L, Smith L, Wagner DL, Blossey B, Brancatini V, Casagrande R, Causton CE, Coetzee JA, Cuda J, Ding J, Fowler SV, Frank JH, Fuester R, Goolsby J, Grodowitz M, Heard TA, Hill MP, Hoffmann JH, Huber J, Julien M, Kairo MTK, Kenis M, Mason P, Medal J, Messing R, Miller R, Moore A, Neuenschwander P, Newman R, Norambuena H, Palmer WA, Pemberton R, Perez Panduro A, Pratt PD, Rayamajhi M, Salom S, Sands D, Schooler S, Schwarzländer M, Sheppard A, Shaw R, Tipping PW, van Klinken RD. 2010. Classical biological control for the protection of natural ecosystems. *Biol. Control* 54:S2–S33.
- van Wilgen BW, Wit MPD, Anderson HJ, Maitre D, Kotzé I, Ndala S, Brown B, Rapholo MB. 2004. Costs and benefits of biological control of invasive alien plants: Case studies from South Africa. *S. Afr. J. Sci.* 100:113–122.
- Wagner WL, Hoch PC. 2005. Onagraceae. The Evening Primrose Family website. Smithsonian National Museum of Natural History, Washington, DC. <http://botany.si.edu/onagraceae/index.cfm>. Accessed August 31, 2019.
- Wagner WL, Hoch PC, Raven PH. 2007. Revised classification of the Onagraceae. *Syst. Bot. Monogr.* 83:1–240.
- Wainger LA, Harms NE, Magen C, Liang D, Nesslage GM, McMurray AM, Cofrancesco AF. 2018. Evidence-based economic analysis demonstrates that ecosystem service benefits of water hyacinth management greatly exceed research and control costs. *PeerJ* 6:e4824.
- Wang H, Moore MJ, Soltis PS, Bell CD, Brockington SF, Alexandre R, Davis CC, Latvis M, Manchester SR, Soltis DE. 2009. Rosid radiation and the rapid rise of angiosperm-dominated forest. *Proc. Natl. Acad. Sci. U. S. A.* 106:3853–3858.
- Wapshere AJ. 1974. A strategy for evaluating the safety of organisms for biological weed control. *Ann. Appl. Biol.* 77:20–211.
- Wheeler GS, Pemberton RW. 2007. A biological control feasibility study of the invasive weed-air potato, *Dioscorea bulbifera* L. (Dioscoreaceae): An effort to increase biological control transparency and safety. *Nat. Areas J.* 27:269–279.
- Wilson JRU, Obinna A, Center TD, Hill MP, Julien MH, Katagira FF, Neuenschwander P, Njoka SW, Ogwang J, Reeder RH, Van T. 2007. The decline of water hyacinth on Lake Victoria was due to biological control by *Neochetina* spp. *Aquat. Bot.* 87:90–93.
- [WSNWCB] Washington State Noxious Weed Control Board. 2019. Noxious Weeds List. Olympia, Washington. <https://www.nwcb.wa.gov/about-the-washington-state-noxious-weed-control-board>. Accessed April 18, 2019.
- Zardini EM, Hongya G, Raven PH. 1991a. On the separation of two species within the *Ludwigia uruguayensis* complex (Onagraceae). *Syst. Bot.* 16:242–244.
- Zardini EM, Peng C, Hoch PC. 1991b. Chromosome numbers in *Ludwigia* sect. *Oligospermum* and sect. *Oocarpon* (Onagraceae). *Taxon* 40:221–230.
- Zardini EM, Raven PH. 1992. A new section of *Ludwigia* (Onagraceae) with a key to the sections of the genus. *Syst. Bot.* 17:481–485.