ASPECTS OF THE INVASION AND MANAGEMENT OF JAPANESE CLIMBING FERN (*Lygodium japonicum*) IN SOUTHEASTERN FORESTS

By

ANDREA N. VAN LOAN

A THESIS PRESENTED TO THE GRADUATE SCHOOL
OF THE UNIVERSITY OF FLORIDA IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF
MASTER OF SCIENCE

UNIVERSITY OF FLORIDA

2006

To my mother and father, for leading by example, and to my husband for his partnership

ACKNOWLEDGMENTS

I would like to express my sincere appreciation to my graduate committee members, who have provided the support and freedom necessary for me to successfully balance my professional, academic, and personal commitments, all of which have been major components of a life-changing period of time spent as a graduate student at the University. First, I thank Dr. Jarek Nowak, who initially chaired my graduate committee, and who recognized the real priorities in my life and allowed me time to devote to them. Dr. Eric Jokela, who began as a committee member, and finished as my committee chair; and gave me the necessary support to move me to completion at a very critical stage. Dr. Greg MacDonald, Agronomy Department representative, has consistently provided friendship, support, and availability at short notice. Dr. Doria Gordon, who provided review, comment, and ecological perspective to my efforts, as well as a dose of reality when stumbling blocks revealed themselves. And finally, Dr Alan Long, who graciously agreed to join my committee very late in the process. Invaluable support was provided by other personnel at the School of Forest Resources and Conservation, including Cherie Arias, and Dr. Wendell Cropper.

This process would not have been possible without the personal and professional support and flexibility provided by my co-workers, especially those within the Florida Division of Forestry's Forest Health Section, and the Florida Division of Plant Industry in Gainesville. In particular, my supervisor, Dr. Ed Barnard, who demonstrated how to truly support staff advancement in consistently approving my flexible work schedules, and leave requests made for field research and academic needs.

The field component of this research would not have been possible without the interest and assistance of several individuals and entities. Critical support for the comparative herbicide trials was provided by Jerry Wyrick, and Wyrick and Sons Pine Straw, and Mr. Elton Headings. Key support for the comparative invasion assessment was provided by: Mr. Dan Miller; the Northwest Florida Water Management District with assistance from Mr. John Valenta; and the Florida Division of Forestry, Blackwater River State Forest with assistance from Mr. Tom Cathey, and Mr. Henry Thompson. This work would not have been possible without support from two funding sources. Research on the impact of site environment on Japanese climbing fern invasion levels was supported by a grant from the University of Florida, Institute of Food and Agricultural Sciences, Center for Aquatic and Invasive Plants, Invasive Plant Working Group. Herbicide research was supported by a grant from the USDA Forest Service Special Technology Development Program (Project # 03-DG-11083112-040).

The role and importance of my family cannot be understated as in the end, all other aspects of life paled in comparison over the last few years. My husband and partner, Chris Van Loan, supported me both at home, and as my research assistant for a significant portion of my field work; providing much-needed labor and assistance despite extreme temperatures, swarming mosquitos, and the relative un-attractiveness of weekends spent in Calhoun County. My mother, Barbara Christman, showed me ultimate perseverance and grace throughout her life. That lesson in perseverance became a touchstone for the writing of this thesis. My father, Harry Christman, showed me the

strength of love as a caregiver in so many senses of the word; allowing me enough peace of mind to continue to make forward progress with this process. While I already see the intrinsic rewards of the academic and research training I have received, these human lessons have been the most humbling and significant of this experience.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Abstract of Thesis Presented to the Graduate School
of the University of Florida in Partial Fulfillment of the
Requirements for the Degree of Master of Science

ASPECTS OF THE INVASION AND MANAGEMENT OF JAPANESE CLIMBING FERN (*Lygodium japonicum)* IN SOUTHEASTERN FORESTS

By

Andrea N. Van Loan

December 2006

Chair: Eric J. Jokela
Major Department: Forest Resources and Conservation

The continual introduction of new non-native invasive species, and the expanding distributions of those already present in the United States make the need for objective priority-based management increasingly important. Japanese climbing fern *Lygodium japonicum* (Thunb.)Sw.) is a non-native weed of mesic to hydric natural communities and sites in the Southeastern United States. Growth ranges from scattered creeping stems to tangled mats with dense canopy, effectively eliminating understory vegetation, and smothering seedlings of overstory tree species. Two field studies explored key aspects of Japanese climbing fern management in forested settings: an assessment of herbicide efficacy for treatment of existing populations, and a review of site characteristics associated with varying levels of establishment.

The assessment of site characteristics associated with varying levels of Japanese climbing fern establishment was conducted in three forested sites in North Florida. Sites were characterized through a comparison of Japanese climbing fern percent cover (dependent variable) with a series of independent plot abiotic and biotic variables; including measurements of soil, and vegetation, assessed on a grid of sampling plots. At

each level of analysis, some plot variables and some species in the plant community showed significant relationships to Japanese climbing fern occurrence levels. The general group of significant variables for presence or absence of Japanese climbing fern included: tree evenness index, plot percent flooded (each winter), soil calcium, and the occurrence of *Carpinus caroliniana*. Comparison of plots with Japanese climbing fern present for varying cover class levels identified photosynthetically active radiation, basal area, soil aluminum, and the occurrence of *Carpinus caroliniana* and *Quercus nigra*. Site variables showing the strongest relationship to Japanese climbing fern occurrence have a unifying basis in site hydrology.

The herbicide treatment study was conducted in a heavily infested forest stand North Florida, where fifteen different herbicides and rate combinations were assessed for effects on Japanese climbing fern foliar cover, compared with an untreated control. Treatments including the herbicide active ingredients glyphosate, imazapyr, and metsulfuron methyl showed the greatest level of Japanese climbing fern control at ten months after treatment; though only glyphosate and imazapyr sustained greater than 70% control to twelve months after treatment.

CHAPTER 1
LITERATURE REVIEW: BIOLOGY, ECOLOGY, DISPERSAL, ECONOMIC IMPACT, AND MANAGEMENT OF JAPANESE CLIMBING FERN

Introduction

“The first time I saw a climbing fern I was shocked, as if I’d opened a window and a trout had flown in” (Hipps 1989).

These words may describe the reactions of many observing the non-native invasive climbing ferns in the southeastern United States. This has been particularly true for Old World climbing fern (*Lygodium microphyllum* (Cav.) R. Br.), as the species has spread rapidly and dramatically across South and now Central Florida. Japanese climbing fern (*Lygodium japonicum* (Thunb.)Sw.) (Family, Schizeaceae), however, has spread with perhaps equal rapidity, through the understory of mesic to hydric sites across Florida. Currently, it is found in nine southeastern states. Many aspects of the life history of Japanese climbing fern indicate the potential for this species to be more broadly pervasive regionally; though perhaps slightly less aggressive on individual sites.

In Florida, Japanese climbing fern establishes dense mats of vegetation reaching 100% groundcover in some areas. Infestations of the plant affect pinestraw management on sites throughout North Florida, and threaten native species and habitats throughout the State. Though increasingly recognized as a pest plant within its established range in the Southeast, Japanese climbing fern may pose a lower threat to agriculture and biodiversity than Old World climbing fern (Pemberton and Ferriter 1998). As a result, greater research focus has been placed on Old World climbing fern, and many gaps exist in the literature on ecology and management of Japanese climbing fern. A review of the literature on Japanese climbing fern follows, and subsequent chapters will address climbing fern management and site ecology. In many areas, references and descriptions of Old World climbing fern are utilized to provide a more comprehensive view of general climbing fern biology and ecology. The known similarities between Japanese climbing fern and Old World climbing fern suggest that a greater understanding of both species can be gained by study of either.

Biology

Rhizomes

Japanese climbing fern rhizomes are widely creeping, bearing leaves in two rows on the dorsal side at the rate of three to four every 1 to 2 cm (Mueller 1982). Rhizomes grow 1 to 3 cm below the soil surface and are densely covered with multicellular hairs (Mueller 1982) which may help to prevent water loss. Roots are numerous, arising endogenously on the lateral and ventral sides near the shoot apex (Mueller 1982). Rhizomes branch dichotomously at 1 to 2 cm intervals after production of three to four leaves (Mueller 1982). As seen in some fern species, activation of rhizoid elongation may be due to presence of key soil minerals (Ko 2003). No examples of vegetative reproduction exist in the literature beyond mention of establishment following transport of intact fronds with rhizomes contained in soil. Rhizomes are easily divided and transplanted due to their creeping, branching nature (Hoshizaki and Moran 2001).

Leaves

Japanese climbing fern leaves are borne on the upper surface of slender, dark brown rhizomes. The rachis of new climbing fern leaves displays “searcher shoot” morphology, similar to many twining flowering plants, with rapid early rachis elongation and delayed leaflet expansion (Mueller 1982). Similar to climbing angiosperms, leaves maintain continual apical growth, exhibit apical dominance (Punetha 1987), circumnutate, and have bud-like dormant meristems on each leaflet. These dormant meristems are capable of growing out to replace a damaged leaf tip, or to strengthen the leaf (Mueller 1981, Mueller 1983).

Japanese climbing fern plants remain evergreen below the frost line in Central and South Florida (Ferriter 2001, Valenta, Zeller, and Leslie. 2001, Zeller and Leslie 2004). Above the frost line, winter dieback of Japanese climbing fern fronds occurs to varying extents through the majority of its range. In the spring, the plants will re-sprout from cold-tolerant subterranean rhizomes (new stems observed the first week in March in North Florida), and often utilize freeze-damaged stems as ladders to grow back into the canopy.

After emergence from the soil in March through May, initial leaf growth is slow (1.2 to 1.5 cm every 3 days), but steadily increases for at least 70 days (Punetha 2000). A series of eight to twelve primary leaves are produced in the first five to six months, before adult leaves are initiated. Successively formed leaves increase in size and take a pinnate form (Mueller 1982). At peak expansion, leaves grow at a rapid rate (3.4 to 6.5 cm per day) for several weeks, including main rachis extension and steady addition of 10 to 25 new alternate primary rachis branches (pinnae). New pinnae emerge from the crozier every 1.7 days (Mueller 1983). Rachis internodes can reach 9 to 10 cm in mature plants (Mueller 1983, Punetha 2000). Leaves exhibit indeterminate growth and can reach lengths of 2 to 30 m (Mueller 1981, Ferriter 2001).

Pinnae on lower rachis are sterile, while successively newer, upper pinnae are increasingly fertile as the growing season progresses. Pinnules (leaflets) are initially tightly coiled, and take five to seven weeks to fully expand to 23 to 25 cm (Mueller 1983). Fertile pinnules bear two rows of sporangia along the underside of the leaflet margin (Ferriter 2001). Leaflets are highly dissected, with a lacy appearance and triangular outline, generally 10 to 20 cm long, and about as wide (Nauman 1993, Langeland and Burks 1998). Leaflet shape and appearance varies noticeably according to age, season, and site. Upon achieving a height of 20 to 40 cm, leaves circumnutate at the rate of ½ to 1¼ rotations per day, typically in a counterclockwise direction (Mueller 1983). Leaves will grow vertically, using trees, shrubs, or other structures for support, or horizontally along the ground (Ferriter 2001), and the climbing habit is undoubtedly an adaptation to increase sunlight access (Holttum 1959). Growth of the stem and leaflets is retarded when the fern is grown either upside down or horizontally (Punetha 2000). Dormant apices on primary rachis branches will proliferate seven days after detopping the frond or delaminating the secondary rachis branches (Punetha 1987, Punetha 2000). When grown under stress (modified habit), the height and vigor of Japanese climbing fern leaves declined significantly (Punetha 2000). In areas with seasonal frost events, Japanese climbing fern foliage will decrease in percent cover during the cooling temperatures preceding the frost event. After frost, Japanese climbing fern foliage turns a rusty-brown color due to stem and leaf death. New leaves emerge from the rhizomes the following March. This seasonal dieback in sub-temperate and temperate climates is one factor which has likely prevented formation of the canopy-covering arboreal blankets seen with Old World climbing fern (Zeller and Leslie 2004). However, the increasing distribution of Japanese climbing fern in south Florida may lead to the development of such canopy-smothering growth on infested sites.

Spores

Japanese climbing fern spores are tetrahedral to globose, with a verrucose surface and trilete aperture that are rust or tan colored. Spores form on the underside of fertile leaf margins which roll under to cover the sporangia, forming false indusia (Langeland and Burks 1998). Japanese climbing fern plants can produce spores year-round in frost-free areas as seen in south Florida (Lott 2001). The effects of seasonal frost and freeze damage can limit much spore production to June through November to December (Lott and Volin 2001) in areas further north. In North Florida, peak spore release apparently occurs in October, when a brown haze of spores is visible in heavily infested stands (Van Loan 2006). Fertile leaflets of Old World climbing fern each produce an estimated 28,600 spores, or 38,000 spores per square inch of fertile leaf area, translating to as many as 30 billion spores on heavily infested sites (Volin, Lott, Muss, and Owen 2004). The typically larger fertile leaflets of Japanese climbing fern may produce a similar quantity of spores (Lockhart 2006).

Spore viability. Once released from the sporangia, spores require moist environmental conditions for germination and survival of the prothallia (gametophyte stage). Spores of *Lygodium* species are thick walled, which extends their environmental viability, an important consideration for management (Ferriter 2001). However, Lloyd and Klekowski (1970) predict short spore viability due to the chlorophyllous nature of the spores. In laboratory conditions, spore germination began within six days of sowing (Lott, Volin, Pemberton, and Austin 2003). Japanese climbing fern spore germination is induced by red light through a phytochrome-mediated system, and is greatly influenced by temperature. Germination rates increase from a low at 20° C as temperatures increase (Kagawa and Sugai 1991, Sahi and Singh 1994), indicating that cooler temperatures will limit both germination and growing season. Sugai, Takeno, and Furuya (1997) found 40% to 80% germination rates for Japanese climbing fern spores after storage at room temperature for 3 years. Perez-Garcia, Orozco-Segovia, and Riba. (1994) found spore viability of *Lygodium heterodoxum* in Mexico drastically reduced following storage in dry conditions (1% germination after 1 year). In contrast, when stored fully imbibed in water germination averaged 40% after 1 year. Light quality was not significant to spore germination, explaining germination success under tree canopies.

Spore dispersal. The spores of Japanese climbing fern are tiny structures, easily transported by wind, water, animals, and equipment (Langeland and Burks 1998), and may possibly travel several miles from their point of origin. The climbing growth form of Japanese climbing fern may also contribute to rapid dispersal by placing spores at the height of the prevailing winds (Lott et al. 2003). Annual flooding in the Appalachicola River floodplain, as well as periodic flooding of other rivers and streams in Florida, distributes Japanese climbing fern spores and frond fragments into new areas (Valenta et al 2001). Land managers have noted observations of increased distribution of the fern in forest stands in the years following the El Niňo-induced flooding events, which occurred across central and north Florida in the winter of 1997/98. These observations may be based on increased recognition of the species, increased distribution of the species due to either flood-based distribution of the spores, or increased soil moisture levels that create more conducive conditions for germination and survival of spores already present in the soil and leaf litter substrate (Ferriter 2001).

The apparent spread pattern of Old World climbing fern in south Florida overlaps with the expected spread pattern if dispersal was primarily due to the prevailing winds of spring, summer and autumn. Old World climbing fern spore production appears to occur year-round, and one measurement of spore loading in the air captured 724 spores per m^3 per hour within a Martin County infestation (Pemberton and Ferriter 1998). Similar, though potentially diminished, spore loading and wind-driven dispersal of Japanese climbing fern may exist given similarities in biology between the two species, although estimates of spore loading are rare for this species in Florida. Some concern exists over the long distance spread of *Lygodium* spores in the convective columns of prescribed or wildfires. These fire-generated wind conditions can disperse smoke great distance away from a fire (more than 100 miles in some cases) and may show the ability to disperse spores in the same way. In addition, soil disturbances associated with forest management activities have been shown to facilitate establishment of the native fern *Dennstaedtia punctilobula* (Michx.) Moore. (Groninger and McCormick 1992), and evidence of a similar effect has been seen with establishment of Japanese climbing fern in some harvested sites in northwest Florida.

Spore banks. Natural and cultivated soils can contain a large number of fern spores in a spore bank, often without close proximity of sporophytes (Hamilton 1988, Dyer and Lindsay 1992, Ranal 2003). When peak spore maturation periods occur, spores may fall to the ground in large, scattered masses; a portion of which percolate into the soil pore space with germination inhibited by lack of light, and others may remain ungerminated on the soil surface (Ko 2003). Unlike limitations on fungal spore germination, fern spore germination does not seem to be affected by soil microbiostasis

(Ko 2003). Ko (2003) suggested that fern spores are nutritionally independent, and germinate successfully on substrates without exogenous nutrients. Soil spore banks may play an important role in reproductive and establishment success for many fern species, creating immediate opportunities for spore germination and gametophyte establishment after any soil disturbance (Ranal 2003). Evidence of fern spore survival in soils for one year has been obtained in multiple studies, and at least one study verifies survival of approximately two-thirds of fern spores for two years (Dyer and Lindsay 1992). Dyer and Lindsay (1992) found that fern spore banks can even survive forest and heath fires. Bark spore banks have been assessed for two tropical species, verifying the potential for such a spore bank to contribute to the population dynamic of pteridophyte species (Ranal 2004). Spores present on bark of live or downed trees, and even in bark products harvested from infested stands may germinate and contribute to dispersal of this species. When coupled with movement of Japanese climbing fern spores in pine straw bales, concerns arise over the possible role of timber and non-timber forest product movement in spore dispersal in the southeastern United States.

Gametophytes/Mating System

Japanese climbing fern is a homosporous fern with free-living haploid gametophytes and diploid sporophytes. Sexually mature gametophytes appear five weeks after spore germination (Lott et al. 2003). In the gametophyte generation, the fern is capable of three types of sexual reproduction: intragametophytic selfing (egg and sperm from same gametophyte), intergametophytic selfing (egg and sperm from two gametophytes originating from the same sporophyte), and intergametophytic crossing (outcrossing of egg and sperm from two gametophytes, originating from two different sporophytes) (Lott et al. 2003). According to Lott et al. (2003) the populations of

Japanese climbing fern in Florida may have originated from only a few plants, with selection favoring selfing variants rather than outcrossing. It has been proposed that Japanese climbing fern has evolved antheridiogens such as gibberellin A_{73}-methyl ester (GA-73-Me) (Wynne, Mander, Goto, Yamane, and Omori 1998), and gibberellin A_9 methyl ester (GA9-Me) (Takeno and Furuya 1980) which may promote outcrossing to avoid the potential inbreeding associated with intragametophytic selfing (Ferriter 2001). However, Lott et al. (2003) found intragametophytic selfing to be the primary mode of reproduction for Japanese climbing fern from two populations in Florida. Over 90% of the gametophytes produced this way developed sporophytes. Therefore, though some evidence of outcrossing exists, intragametophytic selfing may be a stable mating system in Florida. If so, this reproductive strategy in Japanese climbing fern may facilitate colonization of a diversity of habitats and establishment of distant populations from single spores (Lott et al. 2003).

There are four pairs of *Lygodium* species which have some amount of intergrade. This may be due to environmental conditions, or it could be due to hybridization. Japanese climbing fern (*L. japonicum*) has been documented to intergrade with *Lygodium flexuosum* (L.) Sw. (Holttum 1959).

Sporophytes

Under stressful and variable environmental conditions as seen with drought and flood cycles, gametophytes that reach sexual maturity quickly have the best opportunity to produce sporophytes before dying (Lott et al. 2003). Once fertilized in the gametophyte stage, sporophyte production begins. Sporophyte production occurred from week five to week twelve (Lott et al. 2003). Adult plants with climbing leaves and the capability to reproduce have formed by five to six months after spore germination (Mueller 1982) (Figure 1-1).

Ecology

Native Range

Japanese climbing fern is native to temperate and tropical eastern Asia and the East Indies, including India, China, Japan, Korea, and many other countries in the region (Figure 1-2) (Holttum 1959, Singh and Panigrahi 1984, Wee and Chua 1988, Ferriter 2001, Louisiana State University 2006, Royal Botanic Gardens Melbourne 2006, USDA 2006). In its native range, Japanese climbing fern occurs in forest edges, open forests, and secondary forests at both lower and higher (2550 m) elevations, and is described as “never colonizing in any area”(Holttum 1959, Gias Uddin, Mahal, and Pasha 1997); but has also been considered weedy in Taiwan and the Phillippines (Singh and Panigrahi 1984, Langeland and Burks 1998, Ferriter 2001). Holttum (1959) described Japanese climbing fern as “Only found native in regions with pronounced dry season, during which fronds perhaps die.”

Introduced Range

Considered to have been introduced to the United States around 1900 for ornamental purposes, the first naturalized population of Japanese climbing fern was identified in Georgia in 1903 (Pemberton and Ferriter 1998), and in north Florida in 1932 (Gordon and Thomas 1997). Noted as a rare escape in Georgia in 1964 (Radford et al. 1964), it was described as aggressively spreading in moist woods and fields throughout Florida and adjacent states by 1968 (Beckner 1968). In 2005, Japanese climbing fern was identified as “spreading at an alarming rate” in Georgia (Evans and Moorhead 2005). Florida county records for this species have increased from 29 in 1995 to 45 in 1999

(Ferriter 2001), and 53 in 2006 (Van Loan 2006) due to rapid and successful spore dispersal, and improved recognition and reporting. In Florida, recent occurrences in Broward, Collier, and Dade counties as well as Brevard, Hardee, Highlands, Manatee, Polk, and Sarasota counties verify a range overlap with Old World climbing fern (Ferriter 2001, Ferriter and Pernas 2006, Van Loan 2006). This raises concerns that Japanese climbing fern may invade mesic sites and transcend into hydric sites occupied by Old World climbing fern; a situation which is beginning to occur in central and south Florida (Ferriter and Pernas 2006, Lockhart 2006).

As may be true with Old World climbing fern, multiple naturalization events are possible for Japanese climbing fern (Ferriter 2001) though possibly from the same propagative parent plant material (Lott et al. 2003). These two fern species were often confused in early horticultural literature. A photograph and descriptive statements of Old World climbing fern in the 1905 catalog from Royal Palm Nursery (a primary distributor of tropical and subtropical flora in Florida and the United States) actually appear to be Japanese climbing fern. If so, it may have been sold by the nursery for some part of the period (1888-1930) that the nursery marketed Old World climbing fern (Pemberton and Ferriter 1998). Promoted in the horticultural trade as a hardy, attractive plant (Ferriter 2001), Japanese climbing fern is often misidentified as *Lygodium scandens* (L.)Sw., a synonym for Old World climbing fern (Hoshizaki and Moran 2001).

In the United States, Japanese climbing fern now occurs in eleven states including: Florida, Georgia, Alabama, Mississippi, Louisiana, Arkansas, Texas, South Carolina, North Carolina (Figure 1-3), as well as California, Hawaii and Puerto Rico (Figure 1-2) (Ferriter 2001, Van Loan 2006). County-level occurrence records in the Southeastern

United States indicate a close and expanding range overlap with the Coastal Plain physiographic region (Sundell 1986, Martin, Boyce and Echternacht 1993). The Coastal Plain boundary may well serve as a good predictor of the region most rapidly occupied by Japanese climbing fern. Within that range, Japanese climbing fern occurs primarily in floodplain forests, marshes, wetlands, secondary woods, moist pinelands (including flatwoods), and disturbed areas such as culverts, fencelines, road shoulders and rights-of-way (Clewell 1982, Nauman, 1993, Wunderlin 1998, Langeland and Burks 1998). Growth ranges from scattered creeping stems to tangled mats with dense canopy, effectively eliminating understory vegetation (Nauman 1993), and smothering seedlings of overstory tree species (Langeland and Burks 1998). In the Apalachicola river basin, Japanese climbing fern is described as occurring at epidemic proportions in bottomland forests, floodplain forests, sloughs, and mesic flatwoods (Ferriter 2001). Floodplain swamps are comparatively uninfested due to lower elevations and resultant regular flooding and inundation (Ferriter 2001). Infestations also occur in xeric sites, but do not appear to expand as rapidly as in more mesic sites, possibly due to the infrequency of appropriate conditions for gametophyte establishment or fertilization.

Environmental Requirements

The most successful invasive plants are usually capable of displacing native species because they have effective reproductive and dispersal mechanisms, superior competitive ability, limited herbivores or pathogens, ability to occupy a vacant niche, and ability to alter site resource availability, disturbance regimes, or both (Gordon 1998). While the ecological requirements of Japanese climbing fern are poorly understood, its broad distribution indicates a tolerance for a range of environmental conditions. Observations of land managers indicate reduced success in extremely dry or consistently flooded sites (Ferriter 2001) and a potential preference for soils with circumneutral pH (Nauman 1993). Fluctuating precipitation levels may have limited impact on plant growth, but foresters in north Florida reported that in the dry spring of 1999 Japanese climbing fern was not as serious of a problem as in previous wetter springs (Ferriter 2001).

Japanese climbing fern is the most widely established non-native, invasive pest plant on Suwannee River Water Management District lands, and is considered to pose the greatest threat to natural systems on those lands (Ferriter 2001). Twenty populations were evaluated on District lands, and found to occur in the following natural community types: mixed hardwood and pine (9), bottomland hardwood (7), longleaf pine-turkey oak (5), upland hardwood hammock (2), wet hardwood hammock (1), swamp hardwoods (1), and North Florida flatwoods (1). Sixteen of the 20 populations occurred between 50 and 60 feet in elevation. Eighteen of the populations were associated with disturbance (potentially an artifact of the location of the surveys) (Ferriter 2001).

Human disturbance does not appear to be a prerequisite for successful invasion (Ferriter 2001), but in at least two cases in north Florida, removal of pine litter from the forest floor of pine plantations was followed by rapid and complete establishment of Japanese climbing fern in sites where it was previously either absent or present in very small quantities (Van Loan 2001). Sahi and Singh (1994) assessed the effects of sulphite exposure on Japanese climbing fern spore germination and rhizoid growth, and found both to be negatively impacted. This finding indicated that the presence and quantity of Japanese climbing fern gametophytes may be a bioindicator of sulfur dioxide air pollution. This finding may have use in some areas in India, within the native range of Japanese climbing fern.

Japanese climbing fern plants remain evergreen below the frost line, as does Old World climbing fern in central and south Florida (Ferriter 2001, Valenta et al. 2001, Zeller and Leslie 2004). Above the frost line, winter dieback of Japanese climbing fern fronds occurs to varying extents through the majority of its range; plants in shady, moist protected areas like floodplain and bottomland forests, are able to persist through the winter in places. Freeze-damaged/killed fronds turn rusty brown in color and are easily recognized. In the spring, the plants will re-sprout from moderately cold-tolerant rhizomes (new stems observed the first week in March in North Florida), and often utilize freeze-damaged stems as ladders to grow back into the canopy. Japanese climbing fern is promoted as an ornamental plant as far north as Kansas due to cold tolerance of rhizomes, but the Southwestern Fern Society does recommend mulching the plants after the first frost for additional rhizome protection in far northern areas (Ferriter 2001). This seasonal dieback in sub-temperate and temperate climates is one factor which has likely prevented formation of the "dense arboreal blankets in tree canopies seen with Old World climbing fern" (Zeller and Leslie 2004). However, the increasing distribution of Japanese climbing fern in south Florida may soon lead to the development of such canopy-smothering growth on infested sites.

Impacts

In the Apalachicola River basin in Florida, Japanese climbing fern occurs in floodplain forests as a thick mat of growth that smothers native groundcover vegetation, and can extend into the lower overstory canopy (Valenta et al. 2001). Japanese climbing fern is listed as one of the top contributing factors to the degradation of the Apalachicola

River system (Berquist, Pable, Killingsworth, and Silvestri 1995). In its native range, Japanese climbing fern can establish relatively large mats; however, these are much smaller than those commonly found in Florida (Ferriter 2001). The primary effects of Japanese climbing fern on native flora and fauna stem from impacts on understory plant communities, including the habitat of the Florida endangered plant species *Sideroxylon thornei* (Cronq.) Penn., *Aristolochia tomentosa* Sims., and *Polygonum meisnerianum* Cham. & Schlecht. (Ferriter 2001).

Observations of Old World climbing fern, in south Florida and India indicate that the dense growth of fronds into the overstory tree canopy can serve as ladder fuels to carry prescribed fire and wildfire into the canopy, resulting in tree death. Additionally, the fronds serve as a fuel conduit to allow fires to burn into the center of cypress sloughs normally left unburned due to water levels, and embers of the dense fern mats can serve as fuels to carry spot fires away from the primary burn unit (Marthani et al. 1986, Roberts 1996, Roberts 1997). Brandt and Black (2001) found the impact of Old World climbing fern invasion to be a change in vegetative composition of the native community, resulting in a decrease in native species and a loss in plant diversity and evenness. In a comparative study, Clark (2002) found significant reduction in native plant cover, richness and diversity from uninvaded areas to similar habitats with heavy Old World climbing fern infestation. Impacts of a similar type, but possibly lower scale may be seen in sites infested with Japanese climbing fern.

Little is known about the economic impact of Japanese climbing fern invasion. Forest managers in Florida have noted impacts including reduction in annual financial return and harvest lease longevity for pinestraw production, an industry which generates

$79 million in revenue for forest landowners in Florida annually (Hodges, Mulkey, Alavalapati, Carter, and Kiker 2004), and reductions in pine seedling survival during reforestation of heavily infested sites.

Vines as Invasive Plants

An estimated 138 non-native shrub and tree species have invaded native U.S. forest and shrub ecosystems (Pimental, Lach, Zuniga, and Morrison 2004), a figure which does not include invasive grasses, forbs, or vines. However, vines represent a large and aggressive group of invasive plants in the United States. Nineteen of the 133 plant species on the 2005 Florida Exotic Pest Plant Council (FLEPPC) Invasive Plant list are vines, representing 14% of the most invasive plants in Florida (FLEPPC 2005). Horovitz, Pascarella, McMann, Freedman, and Hofstettler (1998) proposed six functional groups of invasive species, including “vine blankets”. The groups were defined to classify recolonization of a site following hurricanes, but other severe disturbance may be considered (i.e. timber harvesting in pine plantations or natural stands). The “vine blanket” group would include both introduced *Lygodium* species in Florida with the proposed effects of shading natives, including: the juvenile/seedling layer, tree re-sprouts from roots and fallen stems, and regrowth of new branches from standing stems.

Vine blanket species are usually aggressive and may negatively influence forest regeneration by diverting resources, and strangling native species seedlings. Vine invasion may drive species composition toward ruderal species, particularly in sites affected by a significant disturbance event (Gordon 1998). Invasion of fire-evolved systems by non-indigenous (and native) vines can alter or prevent fire movement due to increased fuel moisture, and increased site relative humidity. Post-disturbance invasion by non-indigenous vines has been noted in other forests with *Lonicera japonica* Thunb.

Invasion of temperate deciduous forests after smaller-scale natural disturbances have resulted in crushing and strangling of native shrubs and saplings, and contributed to increased root and soil moisture competition (Gordon 1998).

Japanese climbing fern effectively colonizes both basic invasible substrates: those highly disturbed by humans, such as pine plantations, agricultural fields, road shoulders, ditches, and yards; and those relatively free of anthropogenic disturbance such as wetlands, river banks, hardwood hammocks, and some pinelands. Key differences also exist between Japanese climbing fern and several other non-native vines in the Southeast that possess a twining growth form. The twining growth form of Japanese climbing fern contrasts with the bark-adhering growth form of native vines in this area. This twining form enables linkage of tree canopies, increases the likelihood of collapse of the supporting plants, and changes the plant community structure (Hardt 1986, Gordon 1998). Native vines in the Southeast are primarily temperate species, compared with the predominantly tropical origin of the invaders. As a result, the tropical invaders may gain a competitive advantage in the subtropical climates of the Southeast, particularly as seen in Florida (Gordon 1998).

Ferns as Invasive Plants

There are 34 exotic ferns and fern allies in Florida, representing one-third of Florida's fern species (Pemberton 2003). Six of the 133 species on the 2005 FLEPPC plant list are ferns, representing 5% of the most invasive species in Florida (FLEPPC 2005). Pemberton (2003) lists Japanese climbing fern as one of five severely invasive ferns in Florida along with *Lygodium microphyllum, Nephrolepis cordifolia* (L.) Presl. and *N. multiflora* (Roxb.) Jarrett ex Morton, and *Tectaria incisa* Cav., all of which reproduce prolifically and form dense stands or mats of growth, displacing or out-competing native species.

Ferns are also occurring in increasing numbers in the Hawaiian Islands, where 32 alien fern species have become naturalized, including Japanese climbing fern. Much as in the Southeast, the species was not observed to have spread from the original 1936 location until 1998. Since then colonization has expanded rapidly, as additional populations have arisen in multiple locations on O'ahu and Hawai'i (Wilson 2002). According to Wilson (2002) "the Hawaiian ecosystem continues to be challenged by invasion of new species of pteridophytes as well as the spread of already present naturalized species." As a corollary, *Dicranopteris linearis* (Burm.) Underw., a common native climbing fern of Hawaii, is a successful colonizer on open substrates due to factors including: indeterminate (clonal) growth, shallow rhizomes, and a mat-forming capacity (Russell et al. 1998). These same characteristics also contribute to the ability of Japanese climbing fern to colonize habitats in its introduced range.

Related Species in the United States

From 26 to 40 species occur in the genus *Lygodium* (Alston and Holltum 1959, Ferriter 2001) and most are placed in the family Schizaeaceae (Prantl 1881). Two additional *Lygodium* species are established in the Southeastern United States, both of which overlap in range with and create management implications for Japanese climbing fern. A native species, *Lygodium palmatum* (Bernh.) Sw. occurs in swamps, streambeds and ravines in Mississippi, Alabama, and Georgia, and its range extends northeast through New England (Nauman 1993, Miller 2003); while a second, highly aggressive non-native species, *Lygodium microphyllum* (Cav.) R. Brown (Old World climbing fern), occurs in Central and South Florida (Ferriter and Pernas 2006). Native to moist habitats in Africa, southeastern Asia, Australia, and Polynesia (Pemberton 1998, Hoshizaki and Moran 2001), Old World climbing fern has spread rapidly through hydric sites in southern Florida (Pemberton and Ferriter 1998), where the sub-tropical climate, and the fern's life history strategies (climbing vine growth form, highly successful reproductive and long-distance dispersal strategies (i.e., intragametophytic selfing, frond growth into tree canopies, wind and water-driven dispersal), and rapid development of sporophytes within a growing season (Lott et al. 2003)), have facilitated the tendency for invaded sites to be converted to climbing fern monocultures. Old World climbing fern was first observed growing naturalized in Southeast Florida in the 1960s (Beckner 1968, Nauman and Austin 1978). Systematic aerial surveys of Old World climbing fern estimated gross infested area in 1993 at 11,200 ha in South Florida (Pemberton and Ferriter 1998). Similar surveys completed in 2005 increased the estimate of infested area to 48,878 ha in South and Central Florida, a small portion of which may actually be Japanese climbing fern as the species are indistinguishable from the air, and now have overlapping ranges in this area (Ferriter and Pernas 2006). Though Japanese climbing fern is increasingly recognized as a pest plant within its established range in the Southeast, Old World climbing fern has received far greater research emphasis. As a result, much of what is known or published about Japanese climbing fern biology, ecology, impacts, and management is the product or by-product of research and assessment of Old World climbing fern. This relationship has almost certainly enhanced awareness of Japanese climbing fern, but has potentially served to comparatively diminish perceptions of its invasiveness and impact as well.

Recognition and Management in the United States

Management Status

Recognition of Japanese climbing fern has increased among public conservation land managers in the southern United States since the mid 1990s, and has aided in increased detection and reporting. As a result, Japanese climbing fern was designated as a Category I invasive plant by the Florida Exotic Pest Plant Council in 1995, due to it's impacts in "altering native plant communities by displacing native species, changing community structures or ecological functions" (Ferriter 2001, Florida Exotic Pest Plant Council 2005). The State exotic pest plant councils of Alabama, Georgia, and South Carolina also designate Japanese climbing fern as a significant invasive plant (Miller, Chambliss and Bargeron 2004). While non-regulatory, these designations often guide management on public conservation lands. The USDA Forest Service Southern Region has designated Japanese climbing fern as a "Category 1 (exotic plant) species known to be invasive and persistent", guiding it's management on National Forest lands since 2001 (Miller et al. 2004). Recognition of Japanese climbing fern by private land managers has also increased since the late 1990s, but management efforts have not increased at a similar rate, due to lack of awareness of the non-native invasive status of the plant, research-based best management practices, and financial incentives or support for management of private-lands infestations.

Regulatory Status

In 1999, Japanese climbing fern was designated as a noxious weed in Florida (Rule 5B-57.007, F.A.C.) by the Florida Department of Agriculture and Consumer Services (FDACS), making it unlawful to introduce, possess, move, or release any living stage of the fern without a permit. This designation has raised awareness among private forestland managers and members of the forest products industry (esp. pinestraw) in Florida, and for increasing the demand for effective management guidelines. In addition, in Florida, both Collier and Okaloosa counties have local ordinances regulating *L. japonicum* (K. Burks, personal communication, 12/2001). Currently, Alabama is the only other state which designates Japanese climbing fern as a noxious weed (Van Loan 2006).

Japanese climbing fern is not designated as a federal noxious weed according to the Federal Noxious Weed Act of 1974 (7 U.S.C. 2809). A petition was submitted to the United States Department of Agriculture, Animal and Plant Health Inspection Services (APHIS) in 1999, requesting that both Japanese and Old World climbing ferns be given federal noxious weed status. The widespread distribution of Japanese climbing fern excluded it from definition as a quarantine pest, making designation unlikely without further research to more accurately identify the potential distribution in the United States (Ferriter 2001). One potential distribution is shown in Figure 1-3, indicating that much can be done to limit the continued spread of this plant in the United States. Federal noxious weed status would have major impacts on anthropogenic spread of Japanese climbing fern, including: sale of Japanese climbing fern in the horticultural industry, sale of spores in traditional medicine industry, interstate shipment and sale of contaminated forest landscape products.

Economic Impact

In addition to negative environmental impacts, non-native species are causing major economic losses in forestry, agriculture, and other segments of the U.S. economy (Pimental 2004). Little is known about the specific economic impact of Japanese climbing fern invasion. Some estimates place annual economic impact of forest product loss due to invasive species as high as $2 billion in the United States (Pimental et al.

2000). Impacts of invasive plants on the forestry industry in Florida have been estimated at $38 million per year ($15 million in weed control costs, and $23 million in yield losses) (Lee 2005).

Economic costs. Ground-based foliar treatment of Japanese climbing fern in natural areas can range from $45 to $450 per acre, depending on the infestation density, cover and distribution, habitat type, site access and location, and other factors. With multiple treatments required to achieve maintenance control, or eradication if possible, management costs for an infested site can reach the hundreds or thousands of dollars.

Presently, the Florida pine straw industry, which generates $79 million in landowner revenue (Hodges et al. 2004) is most directly impacted by Japanese climbing fern invasion. While quantitative assessment of the economic impact of Japanese climbing fern invasion on the pine straw industry in the southeast has not been conducted, pine straw producers in Florida have noted some impacts. In at least two cases, producers have had to abandon pine plantations leased for straw harvest due to the dense and impenetrable nature of Japanese climbing fern invasion on site and the quantity of economically unacceptable contaminant produced by harvesting infested plantations. Management of this plant in pine straw plantation sites requires additional production cost for the product reducing profits for many producers (Van Loan 2001). In addition, Japanese climbing fern may pose a serious economic risk to pine plantations in the Southeast through spread and intensification of fire in infested areas (Ferriter 2001).

Economic uses. Japanese climbing fern has historically been cultivated in Florida and may have been sold widely for a thirty year period between 1888 and 1930 (Ferriter 2001). Currently it is not being commercially produced in the State (Betrock Information

Systems 2002) due to its noxious weed status. However, sale of Japanese climbing fern continues in other states, facilitated by the internet as in the case of the Bushman Plant Farm of Cleveland Texas (Ferriter 2001).

Japanese climbing fern spores are marketed for multiple herbal treatments under the names spora Lygodii and the Chinese term "hai jin sha" (hai = sea, jin = gold, sha = sand) (Ferriter 2001). Some traditional medicinal uses of spores and spore tinctures include: promotion of diuresis, relief of stranguria (caused by urinary stones), and treatment of eczema (Fan 1996), as well as kidney and urinary function, colds and fever and others (Ferriter 2001). These uses are not expected to be economically significant, but may contribute to a degree of spread of this species.

Management Practices

Integrated pest management utilizes the best appropriate combination of treatments including: chemical, manual/mechanical, cultural, and biological control techniques. Current control technologies for the management of Japanese climbing fern are in development for application in both natural areas and managed properties. At present, herbicide use appears to be the most effective control technique, but further research is need on integration of techniques, and impact of human management practices on anthropogenic spread of the species.

Prevention

With all invasive species, the optimal approach to successful management is prevention of new introductions and prevention of continued spread from existing infestations. At the national level, sale of products containing viable reproductive Japanese climbing fern plant material should be halted, an action which requires establishment of regulatory noxious weed status at the federal level, and throughout the southeastern states. In addition to designation as a noxious weed at the state and federal levels, a strategy for effective enforcement must be identified or developed. The forest products (i.e., timber, pine straw, pine bark mulch) often harvested from infested sites are not easily screened for the presence of Japanese climbing fern spores, but multiple cases in North Florida have verified that in at least some cases, these products are capable of vectoring reproductive material (Van Loan 2006). Further attention to this type of movement will be critical to the success of attempts to manage Japanese climbing fern.

When managing sites infested with Japanese climbing fern, spore vectoring on contaminated personnel, clothing, and equipment must be considered. Hutchinson and Langeland (2006) evaluated Old World climbing fern gametophyte formation from spores collected on clothing and equipment of herbicide applicators after working in three heavily infested sites receiving an initial herbicide treatment, and one site with a very low infestation undergoing a re-treatment. Contamination of clothing and equipment occurred in the initial and re-treatment sites, affecting shirts, pants, sprayers, disposable suits, chainsaws, gloves, machetes and footware. Contamination was significantly greater from the heavily infested initial treatments sites versus the re-treatment sites. Similar results are likely in sites infested with Japanese climbing fern, and applicators and other land managers working in these sites should utilize the following “good spore hygiene recommendations”: spray off equipment and brush off clothing and accessories before leaving site, wash all clothing daily, store disposable suits in plastic bags, limit any vehicle entry into infested areas (Hutchinson and Langeland 2006).

Chemical control

In general, less herbicide testing has been completed on Japanese climbing fern than Old World climbing fern. Initial work that has been completed indicates that glyphosate provides a high level of control. Questions still remain, however, regarding the efficacy of other herbicides, tank mixes, duration of control effect, effects of repeated treatments, and integration of treatment techniques. Preliminary assessments have resulted in a range of herbicide recommendations for Japanese climbing fern management with varying efficacy, including foliar applications of: glyphosate, metsulfuron, triclopyr, and imazapyr (Valenta et al. 2001, Miller 2003, Zeller and Leslie 2004). Valenta et al (2001) evaluated "low", "medium", and "high" rates of glyphosate, triclopyr ester and triclopyr amine each in a replicated study in the panhandle of Florida. At 315 days after treatment (DAT), the glyphosate treatments yielded greater than 70% control (reduction in live foliar cover), while the triclopyr amine and triclopyr ester applications showed no control. In a pair of unreplicated demonstration trials established in north Florida for pine straw producers, Zeller and Leslie (2004) treated inter-rows in two pine plantations, comparing broadcast and spot application techniques, using "high" and "low" rates of glyphosate, triclopyr ester, 2,4-D+dicamba, hexazinone and metsulfuron. When assessed in the period from 300 to 400 DAT, glyphosate broadcast treatments yielded greater than 80% control of Japanese climbing fern, but also caused notable damage to native understory plants. Metsulfuron broadcast treatments showed similar high control levels at 240 DAT, but by 370 DAT, control ranged from 21% at "low" rates to 62% at "high" rates. Damage to native understory plants was lower in metsulfuron treatment areas than observed with glyphosate. In an attempt to begin filling some of the knowledge gaps in Japanese climbing fern management, herbicide evaluations were conducted as a part of this research. The results of these trials, comparing fifteen common forestry and invasive plant herbicide treatments are reported in Chapter 3.

Mechanical control

Mechanical methods for treatment or control of Japanese climbing fern are considered only as components of an integrated management approach, as demonstrated with Old World climbing fern. Observations on Old World climbing fern indicate that vines will re-grow after both cutting of stems and hand-pulling of roots and rhizomes, and that mechanical techniques must be used in conjunction with herbicide application to achieve acceptable treatment efficacy. Stocker, Ferriter, Thayer, Rock, and Smith(1997) assessed a combination of trimming and flooding on Old World climbing fern but found no long-term control. Similar results are likely for Japanese climbing fern. Cutting arboreal fern stems slightly above ground-level prior to herbicide application has been used to increase efficiency of Old World climbing fern treatments in canopy-topping infestations. Heavy equipment has been used in some situations to remove the thick rachis mat formed by the fern's overlapping growth (Ferriter 2001). Use of heavy equipment in climbing fern infestations raises concerns about equipment-based spore vectoring, damage to native species, and exposure of mineral soil aiding re-colonization of sites by Japanese climbing fern or other non-native invasive plants (Hutchinson, Ferriter, Serbesoff-King, Langeland, and Rodgers 2006).

Biological control

Literature searches by Pemberton (1998) revealed very few known insect feeders of Lygodium ferns. However, insect biocontrol candidates have been identified for Old World climbing fern and may exist for Japanese climbing fern. Ongoing research efforts are in place to identify biological control agents for Old World climbing fern with the first approved agent, a pyralid moth, *Austromusotima camptozonale* (Hampson), released in 2005 (Pemberton 2006). It is possible that biological control agents released on Old World climbing fern will have some impact on Japanese climbing fern populations in Florida, particularly below the frost line where populations of the two species overlap, and host preference for Japanese climbing fern is evaluated for all screened agents. The possibility of range overlap with the native *L. palmatum* affects consideration of Japanese climbing fern classical biological control efforts.

A foliar rust fungus (*Puccinia lygodii* (Har.) Arth. (Uredinales)) has been isolated from Japanese climbing fern in Louisiana, and several locations in north and central Florida. The fungus, native to South America, causes severe damage to the leaflets, including necrosis, browning, and drying (Rayachhetry, Pemberton, Smith, and Leahy 2001), and has been observed to apparently increase in distribution and impact in north Florida climbing fern populations. The visual impact of this fungus on climbing fern in north Florida has been most noticeable in early winter (i.e., November and December), when varying levels of damage have been observed on approximately 95% of foliage in some forest stands in north Florida. In preliminary observation, the primary impact of this fungus in north Florida appears to be a reduction in photosynthetically active leaf surface area immediately preceding seasonal leaf dieback. The fungus does not appear to affect fern re-growth in the spring. Rust fungi are considered difficult to propagate in laboratory conditions, possibly limiting further consideration of *P. lygoddii* as a bioherbicide candidate. Strandberg (1999) evaluated the effects of inoculation with the *Colletotrichum acutatum* H. Simmonds (fern anthracnose) fungus, but found disease damage on Japanese climbing fern to be minimal. Jones, Rayamajhi, Pratt, and Van

(2003) evaluated the fungus *Colletotrichum gloeosporioides* (Penz.) Penz. & Sacc. for pathogenicity on both invasive climbing ferns, finding some browning and wilting of the pinnules of Japanese climbing fern, and as much as 50% necrosis on Old World climbing fern plants.

Prescribed fire treatment

Foresters in north central Florida have reported that prescribed fire is not effective in controlling Japanese climbing fern in pine plantations (Ferriter 2001). The effects of prescribed burning have not been directly evaluated on Japanese climbing fern; however, results might be expected to be similar to those in Old World climbing fern. Prescribed fire was evaluated both alone and in combination with herbicides for control of Old World climbing fern by Roberts and Richardson (1994), and Stocker et al. (1997). Roberts and Richardson (1994) concluded that fire alone cannot be used to manage Old World climbing fern based on measurements of percent cover and height and number of stems. Fire used in combination with a single herbicide application was ineffective as well. Stocker et al. (1997) compared combined treatments of trimming and flooding, contact herbicide and flooding, and burning and flooding, and found Old World climbing fern to successfully recover following all treatments. Prescribed fire has been found to be an effective biomass reduction tool prior to herbicide application, resulting in a reduction in herbicide required to control Old World climbing fern (Ferriter 2001).

Haywood et al. (2001) assessed the impacts of 37 years of seasonal prescribed burning on vegetation in a dry-mesic upland longleaf pine site in Louisiana. In measuring occurrence of herbaceous vegetation on the study plots, Japanese climbing fern was found to be the dominant species on 3% of the plots burned biennially in March, and the second most common species on 3% of the plots burned biennially in July.

Japanese climbing fern was less common on plots burned biennially in May. Anecdotal observations on the study plots indicate that regardless of burn regime, Japanese climbing fern is increasing in presence on all plots.

Similar increases in percent cover of Japanese climbing fern on regularly burned sites have been observed in north and central Florida (J. Wyrick, 6/15/2002, personal communication.). Climbing fern infestation may alter fire behavior on a site by serving as a ladder fuel to carry flames into the tree canopy. In addition, concern exists about the possible long-distance spread of climbing fern spores in the convective columns of prescribed fires or wildfires (Ferriter 2001). These fire-generated wind conditions can disperse smoke great distances away from a fire (more than100 miles in some cases) and may show the ability to disperse spores in the same manner.

Conclusion

While some aspects of the biology, ecology, and management of Japanese climbing fern have been described, clear gaps do still exist. Questions remain about invasiveness in various habitats, interactions with native plant species, spore longevity in the environment, effects of forest management practices, anthropogenic spread, and many more. The chapters which follow include further exploration of habitat/invasion associations, and herbicide management in an attempt to address some of these questions.

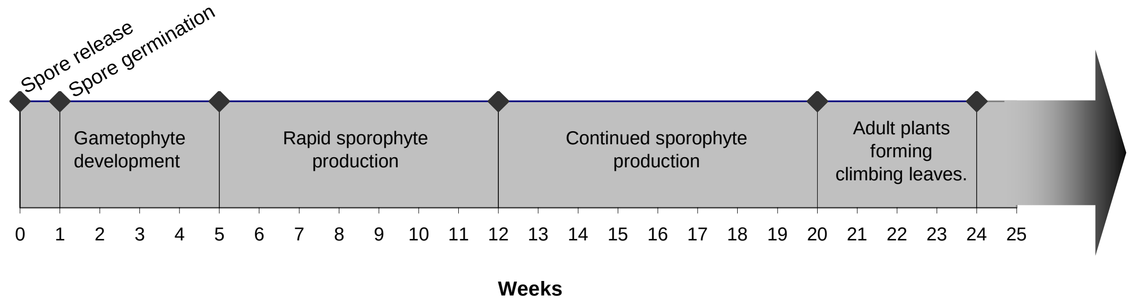

Figure 1-1. Timeline for Japanese climbing fern plant development from spores.

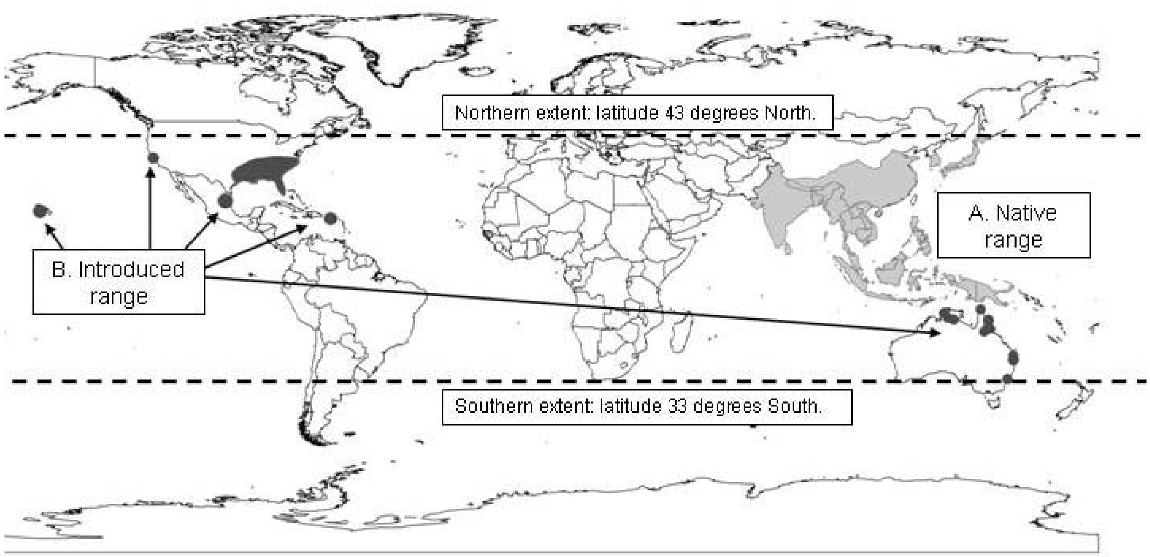

Figure 1-2. Worldwide distribution of Japanese climbing fern (*Lygodium japonicum*) from literature review (June, 2006). A) Native range indicated in grey. B) Introduced range indicated in black. Dotted line indicates northern and southern extent of recorded Japanese climbing fern field populations.

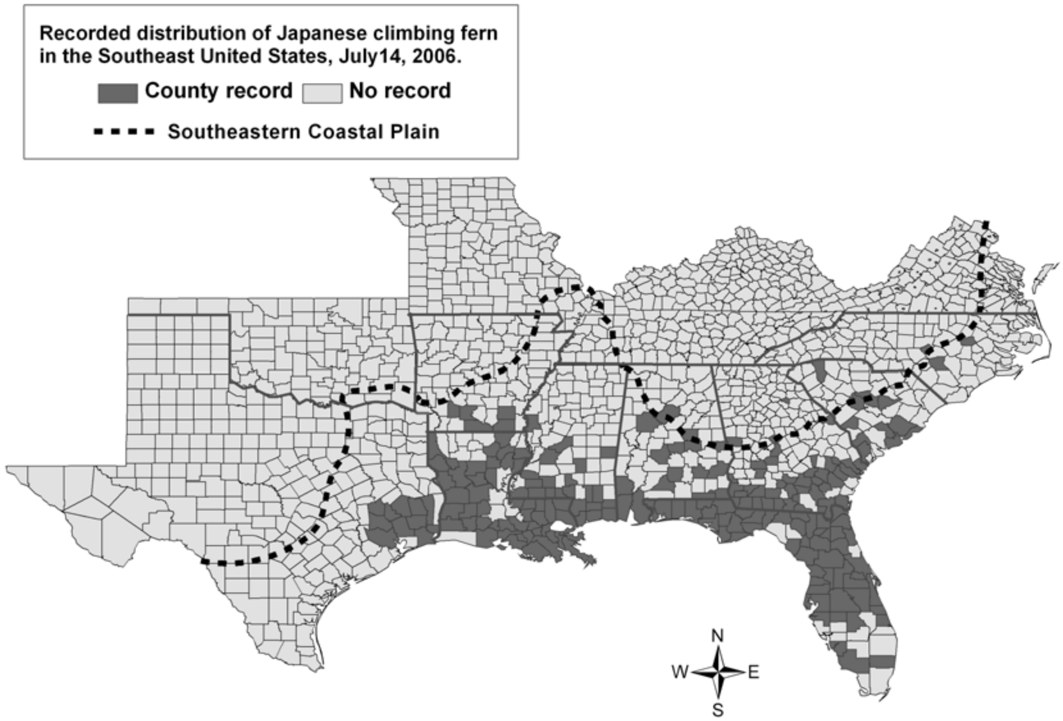

Figure 1-3. County-level distribution of Japanese climbing fern in the Southeastern United States with reference to the Southeastern Coastal Plain physiographic region. Records based on vouchered herbarium specimens from Southeastern herbaria, and records from regional vascular plant databases.

CHAPTER 2
ASSESSMENT OF SITE VARIABLES ASSOCIATED WITH JAPANESE CLIMBING FERN OCCURRENCE IN THREE NORTH FLORIDA FORESTS

Introduction

A simplified view of plant invasions includes two essential stages: transport of organisms or propagules to new sites, and establishment and population expansion in the invaded sites (Shea and Chesson 2002). Both stages are affected at some level by the conditions of the invaded site. On any given site or region, invader growth rate is affected by resources, the physical environment, ecological adaptations of the invasive species, and natural enemies. Invasive potential may be determined by environmental factors including soil pH and nutrient levels, site climatic conditions and native plant community (Langeland and Burks 1998; Davis, Grime, and Thompson. 2000; Shea and Chesson 2002). Thus, the mechanisms that determine site invasibility by non-native species, in various ecosystems, are often unknown (Lavorel, Prieur-Richard, and Grigulis 1999). Japanese climbing fern is one species for which the details establishment success are poorly quantified. Some fern species are dependent on specific site conditions for successful establishment and spread, including climate, soil substrate, soil pH, elevation, and moisture regimes (Peck, Peck and Farrar 1990; Whittier and Moyroud 1993; Stewart 2002). Establishment and persistence of gametophyte populations are strongly affected by environmental events (Peck et al. 1990). Most ferns in the Schizaeaceae are vigorous in edaphically low nutrient environments (Page 2002). This tolerance opens a variety of sites to establishment, including intrinsically nutrient poor savannahs, post wildfire sites, erosion surfaces, and epiphytic habitats (Page 2002).

While the ecological requirements of Japanese climbing fern are poorly understood, its broad distribution indicates a tolerance for a range of environmental conditions. In its native range, Japanese climbing fern occurs in forest edges, open forests, and secondary forests at both lower and higher elevations (Singh and Panigrahi 1984, Gias Uddin et al. 1997, Ferriter 2001). In the Southeast United States, Japanese climbing fern occurs primarily in floodplain forests, marshes, wetlands, secondary woods, moist pinelands (including flatwoods), and disturbed areas such as road shoulders and rights-of-way (Clewell 1982, Nauman, 1993, Wunderlin 1998, Langeland and Burks 1998). Infestations also occur in xeric sites such as sandhills, but do not appear to expand as rapidly as in more mesic sites.

Observations of land managers indicate reduced success in extremely dry or consistently flooded sites (Ferriter 2001) and a potential preference for soils with a pH close to 7 (Nauman 1993). Most ferns flourish at pH between 6 and 7 (Hoshizaki and Moran 2001). Fluctuating water levels may have limited impact on plant growth, but foresters in north Florida reported that in the dry spring of 1999, Japanese climbing fern was not as serious of a problem as in previous wetter springs (Ferriter 2001). Human disturbance does not appear to be a prerequisite for successful invasion (Ferriter 2001), but populations are often associated with ditches and culverts due to increased site moisture levels, and possibly due to associated soil disturbance as well. .

The objectives of this study are to classify and describe biotic and abiotic variables associated with the presence or absence, and percent cover of Japanese climbing fern, as well as those associated with varying levels of percent cover. Identification of clear associations between site environmental factors and fern establishment success may aid prioritization of treatments for managing Japanese climbing fern invasions, and improve understanding of habitat vulnerability to fern invasion.

Methods and Materials

Site Selection

Three forested study sites were selected in North Florida (Figure 2-1), each containing large areas infested with Japanese climbing fern, and no history of herbicide treatment in the past 10 years. Sites were selected in areas with visually “heavy” local regional infestation levels, with the intention of reducing variability in site spore loading, and therefore in propagule-based establishment opportunity. Sites were also selected to maximize natural community variability, with multiple natural community types represented at each study area. Plot measurements were taken between June and September, 2002.

Plot Layout

On each site, a square macroplot 400 m by 400 m in size (16 ha) was identified as the study area and subjected to ecological sampling. Percent cover of Japanese climbing fern was determined through line intercept (Myers and Shelton 1980), with intensive sampling plots distributed in equal distances along transect lines (Figure 2-2). In each study site, a baseline was established along a road marking a stand boundary and linear disturbance, or “edge”. Five parallel major transect lines were established, originating from the baseline at 100 m intervals, and extending 400 m into the stand, perpendicular to the baseline.

A grid of 25 intensive sampling plots (plots), each 10 m by 10 m in size, were established at 100 m spacing along the major transect lines. The center of each plot was permanently marked. On each plot, five minor transect lines were established, each 10 m long and spaced 2.5 m apart. A grid of 25 sampling points (points) was established every 2.5 m along these minor transect lines (Figure 2-2). Each point was marked with a pin flag. Percent cover of Japanese climbing fern was determined for all major transect lines and minor transect lines through line intercept measurements.

Site Descriptions

Site 1. Blackwater River State Forest, Santa Rosa County (BRSF) is a publicly-owned 78,000+ ha property managed by the Florida Division of Forestry. The study area included natural, fire-managed upland sandhill community, with a temporal stream drainageway cutting curvilinearly through a portion of the site (Figure 2-3). A large portion of the drainageway is heavily infested with Chinese privet (*Ligustrum sinense* Lour).

Site 2. The Miller property, in Calhoun County (Miller) is a privately owned forestland with portions of the study area in the following community types: mesic flatwood slash pine plantation (previously managed for pinestraw production), bottomland hardwoods, longleaf pine upland, mixed pine/hardwoods, and cypress wetlands (Figure 2-4).

Site 3. Florida River Island, in Liberty County (FRI) is a publicly-owned floodplain forest island situated between the Apalachicola and Florida Rivers, and managed by the Northwest Florida Water Management District. The study area is primarily cypress swamp, and floodplain swamp hardwood forest, including areas of higher elevation on ridges and natural levees with limited soil saturation, and low flat swales which remain inundated for much of the year (Figure 2-5) (Leitman, Sohn and Franklin. 1983).

Macroplot Measurements

Physical characteristics of each site (16 ha macroplot) were assessed, including locally significant natural features (bodies of water, roads, etc.), and natural community boundaries; delineated using field mapping and interpretation of aerial imagery (Figure 2-3, Figure 2-4, Figure 2-5). Percent cover of Japanese climbing fern in the understory was measured using the line-intercept method along the 400 meter-long major transect lines to generate an average percent coverage by site. The site-level values were compared to average plot-level values on each site for significant differences in measured percent cover. Lack of agreement between the two measurements would have indicated an inadequate sampling design. The percent cover of Japanese climbing fern was defined as: the total linear distance covered by the foliage over the entire length of the transect lines within a site, divided by the total transect line length and multiplied by "100" (Myers and Shelton 1980). The same measurements were taken along the 10 meter-long minor transect lines on each plot, and the same technique used to generate an average percent coverage for each intensive sampling plot.

Intensive Sampling Plot Measurements

The majority of the classifying measurements were taken on the 10 m by 10 m intensive sampling plots. For each plot, the following features were measured on each of the 25 sampling points within the plot to generate a composite average value for the plot. Plot-level percent cover of Japanese climbing fern was measured and was the dependent variable for subsequent analyses. The species and diameter at breast height (dbh), was measured for all trees (minimum 2 inch dbh) on the 10 m by 10 m intensive sampling plots. These measurements were used to aid in typification of site, and allow for calculation of plot basal area (m^2 ha^{-1}), species richness, species diversity, species evenness, and species importance values.

Understory vegetation, and midstory vegetation percent cover were recorded as ocular estimates of all plant material excluding Japanese climbing fern. Overstory percent canopy cover was determined visually using a non-spherical densiometer. Values were recorded on a scale with 100% being full canopy with no light penetration, and 0% being no canopy overhead. Relative light penetration ($\mu mol\ m^{-2} sec^{-1}$) was determined by measurements of photosynthetically active radiation (PAR) with a Li-Cor light meter inside the stand and in the open. Average light intensity for each intensive sampling plot was obtained by measuring PAR just above the forest floor vegetation over the 25 sampling points for each plot, and averaging these values to obtain a plot average. These averages were then corrected with a relative PAR factor measured under zero canopy conditions for the respective sampling period to determine the relative light penetration for each intensive sampling plot measured that day. Light readings were taken between 10:00 am and 2:00 pm on days with no cloud cover to ensure uniformly peak light conditions.

Site hydrology was described with three variables: Hydrology class (Xeric=1, Xeric/Mesic=1.5, Mesic=2, Mesic/Hydric=2.5, Hydric=3) assigned according to site observations, percent of the plot flooded from winter 2002 through spring 2003, and the number of months each plot remained flooded. Plot topographic relief was ocularly assessed as concave, convex, or flat. Plot percent slope was determined using a hypsometer. Slope aspect was determined with a compass.

A soil core (diameter 2 cm, depth of 15 cm) was collected with a soil probe at each of the 25 points per plot to generate a composite plot soil sample. Composite soil samples were allowed to air dry, and screened to pass a 2 mm sieve prior to analysis for: macronutrients, organic matter content and soil pH. Mehlich-1 extractable P, K, Ca, Mg, Cu, Mn, Zn, Al, Na, and Fe were measured using the Mehlich-1 extraction solution (0.0125 M H_2SO_4 and 0.05M HCl) to provide a soil solution ratio of 1:4. Units for all elements are recorded as mg per kg. Soil pH was measured using a 2:1 water to soil ratio. Soil organic matter percent was measured through Walkley-Black dichromate methodology. This method was originated to assist in the selection of herbicide rates for mineral soil containing less than 6% organic matter.

Data Analysis

Plot environmental variables and calculated indices were usedto assess relationships between variables and the presence or absence of Japanese climbing fern across all plots. To improve interpretation, results of these analyses are presented in four functional groups: plot abiotic variables, plot soil variables, plot vegetation variables, and tree species importance values. Variables were further assessed for relationships between three levels of Japanese climbing fern percent cover across plots with Japanese climbing fern present.

Plot abiotic variables include: photosynthetically active radiation, percent slope, percent exposed soil, plot hydrology class, plot percent flooded (2003), and months flooded (2003). Plot soil variables include: pH, P, Ca, Fe, Mg, Al, K, Zn, Na, Mn, Cu, and percent organic matter. Plot vegetation variables include: understory percent cover, midstory percent cover, and tree canopy percent cover. Plot tree inventory data were utilized for the calculation of additional plot vegetation variables: tree basal area (m^2/ha), tree species richness (species per plot), Simpson's Reciprocal (1/D) Index (Magurran 1988) of tree species diversity, and Pielou's J' Index of tree species evenness (Pielou 1975).

Tree importance values were calculated for each species on each plot using the equations below:

- IMPORTANCE VALUE: Relative density + relative dominance + relative frequency.
- DENSITY: Total stems of "species"/total plot area.
- RELATIVE DENSITY: (Density of "species"/sum density of all species) X 100.
- DOMINANCE: Mean basal area per tree of "species" X number of trees in species.
- RELATIVE DOMINANCE: (Dominance of "species"/sum dominance of all species) X 100.
- FREQUENCY: (Plots with "species" / Total number of plots) X 100.
- RELATIVE FREQUENCY: (Frequency of "species"/sum frequency of all species) X 100.

Data Transformation. Anderson-Darling test statistics were calculated to assess normality of distributions for each environmental variable, across all study plots (n=75, alpha=0.01). Distributions of most variables were non-normal. Non-normal data were then transformed and normality re-assessed. Natural log, $\log_{10}$, and $\log_2$ transformations were performed on all variables; a constant (value=1) was added to each variable value prior to log transformation to allow inclusion of zero values. Additionally, inverse, and square root transformations were performed on all variables, and arcsine transformations were performed on variables with percentage values. Transformed data showing the strongest probability of normal distribution for each variable were utilized in subsequent parametric analyses of environmental data. Tree importance values were assessed using non-parametric analyses. The data subset including only plots with Japanese climbing fern (n=29) was also assessed for normality of distribution. In this case most variables had normal distribution of values.

Initial Analyses

Presence/absence. Initial analyses were conducted to measure the significance of differences in plot environmental variables and tree importance values, on the presence or absence of Japanese climbing fern. Significant differences in variable mean values between plots with Japanese climbing fern present versus absent were assessed using the Kruskal-Wallis one-way rank analysis of variance test (alpha=0.05) for non-normally distributed variables. Kruskal-Wallis p-values are calculated using the chi-square approximation (Siegel 1988). Variance equality was assessed for normally distributed plot environmental variables using the folded F-statistic (alpha=0.05). Significant differences in normal variable mean values between plots with Japanese climbing fern present versus absent were assessed using a pooled two-sample t-statistic for variables with equal variance, and Satterthwaite's two-sample t-statistic for those with unequal variances. Significant correlations between variable mean values on plots with Japanese climbing fern present versus absent were assessed using the Spearman R correlation coefficient (alpha=0.05) for non-normal variables, and the Pearson correlation coefficient (alpha=0.05) for normal variables. Correlation strength ratings were assigned based on coefficient values: weak=0.2 to 0.49, moderate=0.5 to 0.79, strong=0.8 to 1.0 (Cohen 1988).

Cover class. Separate analyses were conducted for all plots with Japanese climbing fern present (n=29) from all sites, to assess relationships between three Japanese climbing fern cover classes, and plot environmental variables and tree importance values. Normal distributions were achieved with raw data for most variables, allowing use of parametric tests where appropriate without data transformation. Cover classes utilize the following ranks: 0= 0%, 1= >0 to 5%, 2= >5 to 25%, 3= 26 to 75%, 4= 76 to 95%, 5= 96 to 100% (Horvitz and Koop 2001). All plots fell in cover classes 1, 2, or 3. Parametric one-way analysis of variance tests (alpha=0.05), and non-parametric Kruskal-Wallis analysis of variance tests (alpha=0.05) were performed on all environmental variable and tree importance values by cover class.

Discriminant Analysis

Plot environmental variables and tree species found to vary significantly according to Japanese climbing fern presence or absence, and Japanese climbing fern cover class levels were evaluated in stepwise discriminant analyses of covariance. Analyses were conducted on plots at the study-level (n=75) and site-level (n=25 each). Plot variables and tree values were assessed separately. Significance was determined by the results of the Kruskal-Wallis analysis of variance tests and two-sample t-tests. Power for variable entry or removal in the discrimnant analysis (alpha=0.15) was measured by the Wilk's Lambda likelihood ratio criterion (SAS Version 9.1). Data were assessed at the study-level and the site-level.

Regression Analysis

Multiple logistic regression analysis was performed on significant variables from the Japanese climbing fern presence/absence assessment, and the Japanese climbing fern cover class assessment. Spearman R correlation coefficients were calculated across all variable to assess levels of variable correlation. Principal components analysis was used to combine strongly correlated (correlation coefficient exceeds 0.8) environmental variables into the component factors for use in the regression analyses.

Neural Network Analysis

Artificial neural network analysis was conducted utilizing all plot variables to develop a deterministic classification model to assess the ability of the data set to correctly classify plots according the presence or absence of Japanese climbing fern. The model was developed as a “C” program, and modified for use in Python. Plots were assigned “0” or “1” values according to Japanese climbing fern absence or presence respectively; these values served as the supervised network teacher. A non-linear activation function in the model used a radial basis transfer function, which assumes a Gaussian curve. Network inputs, hidden layers (the radial basis transfer function), and network outputs are connected by weight multipliers, summed by nodes.

Results

Results of respective analyses are presented in Tables 2-1 through 2-10, and appendix tables, and Figures 2-3, 2-4, and 2-5 at the end of this chapter. To improve interpretation, results of initial statistical tests are presented in four functional groups: plot abiotic variables, plot soil variables, plot vegetation variables, and tree species importance values.

Site-Level Occurrence

Percent cover of Japanese climbing fern on all sites is presented in Table 2-1, including comparisons between plot measurements taken in 2002, and re-assessed on two sites in 2004. Among the three sites, Florida River Island (FRI) had the highest site-level and plot-level percent cover of Japanese climbing fern in 2002 (13.7%), and the greatest number of plots infested (18), but it could not be re-assessed in 2004 due to a herbicide treatment applied to the Japanese climbing fern on the site in 2003. The Miller site had the second highest site-level percent cover (3.00%), with eight plots infested in 2002, while percent cover on the Blackwater River state forest site was lowest (1.3%), due to a small number of infested plots (3) with lower individual percent cover levels. The number of plots with Japanese climbing fern present increased from 2002 to 2004 by one plot each on the Miller site and the Blackwater River State Forest (BRSF) site. Plot percent cover of Japanese climbing fern increased (0.4%) from 2002 to 2004 on the Miller site, but decreased (0.6%) on the BRSF site over the same time period. Two-sample t-tests revealed that neither change was significant (alpha=0.05). Plot-level Japanese climbing fern presence and percent cover values from 2002 are used in statistical analyses. Macroplot (site-level) cover was not re-assessed

Japanese climbing fern cover classes are illustrated in Figure 2-3 (BRSF), Figure 2-4 (Miller), and Figure 2-5 (FRI). Across all sites, plots on xeric pine uplands were uninfested, while mesic pine community types (pine plantation, mesic flatwoods, mixed pine/hardwood), and hydric community types (hydric cypress wetland, hydric hardwood swamp, and hydric floodplain forest: ridge, swale, and slough) each averaged a 55% Japanese climbing fern occurrence level. Table 2-2 indicates Japanese climbing fern presence/absence levels according to natural community type.

Plot-Level Occurrence

Presence/Absence

Plot environmental variables. Means of the following plot abiotic and biotic variables varied significantly (alpha=0.05) according to Japanese climbing fern presence (in order of significance): site, PAR, understory percent cover, canopy percent cover, Pielou tree evenness index, hydrology class, Simpson's reciprocal tree diversity index, tree species richness, months flooded, percent flooded, and trees per plot. Understory percent cover and PAR were negatively correlated with Japanese climbing fern presence; all other significant variables showed positive correlations. Correlation strengths were weak (Table 2-4, Tables A-1 to A-4).

In particular, photosynthetically active radiation ranged from 133 μmol $s^{-1}m^{-2}$ on sites with Japanese climbing fern present to 455 μmol $s^{-1}m^{-2}$ on sites without Japanese climbing fern. Hydrology class increased in moisture level with Japanese climbing fern presence. Flood duration averaged two months on sites with Japanese climbing fern compared to one month on sites without Japanese climbing fern, and percent flooded increased from 15% to 27% with the presence of Japanese climbing fern.

Means of the following plot soil variables varied significantly (alpha=0.05) in parametric tests of relationship to Japanese climbing fern presence: pH, phosphorus (P), calcium (Ca), iron (Fe), magnesium (Mg), aluminum (Al), potassium (K), zinc (Zn), and sodium (Na). Manganese (Mn), copper (Cu), and percent organic matter did not vary significantly (Table A-3). Phosphorus and pH were moderately correlated with fern presence, while other significant variables were weakly correlated. Iron and aluminum demonstrated negative relationships with fern presence, while other significant variables were positively correlated. Soil pH on sites with Japanese climbing fern present averaged 6.0, while sites without Japanese climbing fern were slightly more acidic, averaging 5.2. Soil aluminum was significantly higher on plots with Japanese climbing fern absent (332 mg kg^{-1}) than plots with fern present (192 mg kg^{-1}). Conversely, higher levels of soil calcium occurred in plots with Japanese climbing fern present (284 mg kg^{-1}) than absent (101 mg kg^{-1}).

Plot tree species. Importance values were analysed by species, comparing mean values for plots with Japanese climbing fern present versus absent. Forty-three tree species were identified across all plots, of which twenty-one occurred on one plot only. Species with importance values that varied significantly according to Japanese climbing fern presence or absence include: *Liquidambar styraciflua, Pinus palustris, Carya aquatica, Quercus lyrata,* *Ulmus americana, Carpinus caroliniana, Quercus nigra*, and *Taxodium distichum* (Table 2-7, Table A-4), while those not exhibiting a significant relationship are listed in Table A-5. Mean importance values for the majority of significant tree species demonstrate a positive relationship with Japanese climbing fern presence; the one exception being *P. palustris* which demonstrates a moderate negative relationship. Other significant species are, at best, weakly correlated with Japanese climbing fern presence.

Tree species were assigned vegetative index classes according to the Florida Department of Environmental Protection's Wetland Delineation Index (Sec.62-340.450, F.A.C.) (FLDEP 2006). Analyses were conducted to assess relationships between tree importance values within vegetative index classes and Japanese climbing fern presence or absence (Table 2-3). Tree importance values on plots with Japanese climbing fern present versus absent were compared, significant differences between Japanese climbing fern presence and absence in importance values for facultative upland trees, and obligate wetland trees. Analysis of variance detected significant ($P=0.005$) variation in importance values between vegetative index classes on plots with Japanese climbing fern present, as well as on plots with Japanese climbing fern absent ($P<0.0001$).

Cover Class Levels

Plots with Japanese climbing fern present were analyzed as a subgroup to assess relationships between three Japanese climbing fern percent cover levels (raw data or cover class), and plot environmental variables and tree importance values. No significant (alpha=0.05) relationships were detected between plot and tree variables and Japanese climbing fern cover class in parametric one way analysis of variance. However, significant (alpha=0.05) nonparametric relationships were detected between percent cover of Japanese climbing fern and three variables, including plot basal area which peaked (30 m^2 ha^{-1}) on moderately infested (class

2) plots. Soil aluminum reached 249 mg kg^{-1} on moderately infested plots, an average of 70 mg kg^{-1} more than on slightly (class 1) or heavily (class 3) infested plots. Occurrence of *Carpinus caroliniana* also showed significant relationships to Japanese climbing fern cover class. (Table 2-4). Significant (alpha=0.05) correlations to Japanese climbing fern cover class were measured for basal area, plot percent flooded, photosynthetically active radiation, and the occurrence of *Carpinus caroliniana* and *Quercus nigra* (Table 2-4).

Discriminant Analysis

Stepwise discriminant analysis of covariance was used to further explore relationships between plot variables and Japanese climbing fern occurrence, and cover class. Variables were measured for relationships at the study (n=75) level, and at each respective site (n=25). Across all study plots, soil calcium was the most significant discriminator (P<0.0001) of Japanese climbing fern presence or absence (Table 2-5). Site level analyses revealed relationships between other variables including percent flooded (P=0.02), and months flooded (P=0.0001) at BRSF; while soil iron is significant (P=0.002) at the Miller site, and tree species richness is significant (P=0.03) at FRI. (Tables 2-5 and 2-6) Of the variables that discriminated between Japanese climbing fern presence or absence (Table 2-5), percent flooded is the only one also significant (P=0.09) as a discriminator (alpha=0.10) between Japanese climbing fern cover class levels (Table 2-6). In addition, soil aluminum and plot basal area were also both significant, discriminators of Japanese climbing fern cover class at the study level. Site level analyses revealed the strongest relationship between variables and Japanese climbing fern cover class, to be soil aluminum at the Miller site (P=0.01), followed by percent flooded at the Miller site (P=0.14). At FRI, basal area and soil aluminum were significant (alpha=0.15) discriminators of Japanese climbing fern cover class.

Quercus nigra was a significant discriminator between Japanese climbing fern presence and absence at the study level (n=75, P<0.0001) and at BRSF (n=25, P<0.0001) (Table 2-7); as well as between cover class levels at Florida River Island (Table 2-8). Other significant tree species were discriminators of Japanese climbing fern presence or absence, such as: *Pinus palustris* (P<0.0001) and *Liquidambar styraciflua* (P=0.0005) at the study level, *Ligustrum sinense* at BRSF (P<0.0001), *Pinus elliotii* at the Miller site (P=0.02), and *Nyssa aquatica* (P=0.02) and *Acer rubrum* (P=0.08) at FRI. *Carpinus caroliniana* was the only significant discriminator of cover class (P=0.02) at the study level. Discriminant analysis was not possible for BRSF due to the small number of plots with Japanese climbing fern present; and no tree species were accepted into the analysis for the Miller site.

Regression Analysis

Multiple logistic regression analysis was performed on significant variables from the Japanese climbing fern presence/absence and cover class assessments. Principal components analysis was used to combine strongly correlated (correlation coefficient exceeds 0.8) environmental variables into the following factors: FLOOD=Hydrology class + plot percent flooded + months flooded, SOIL1=pH (inverse) + phosphorus (inverse) + calcium (natural log), SOIL2=Zn + K (natural log), TREE=Pielou species evenness index + Simpson's reciprocal species diversity index + species richness. Principal components analysis explained more than 80% of the variance among factor components for all factors, indicating substantial agreement, and validating use of factors in subsequent multiple regression analysis.

Variables generating significant effects in the regression analyses are listed in order of forward selection into the regression models in Tables 2-9 (presence/absence) and 2-10 (cover class). The first entry into the presence/absence model is an intercept (value=0.4), followed by the principal component factor SOIL1, accounting for 40% of the variation in the model.

Following general site classification by those soil pH correlates, the additional cumulative effect of importance values of *Ligustrum sinense, Quercus nigra, Lindera benzoin*, *Acer rubrum*, *Quercus laevis*, and *Nyssa aquatica* accounting for a maximum of 68% of the model variation related to presence or absence of Japanese climbing fern. When the presence/absence analysis was conducted for each respective site, few variables were entered into the model. At BRSF, the principal components analysis factor TREE was entered, accounting for a cumulative 49% of model variation. At the Miller site, the SOIL1 factor entered the model, followed by *Pinus palustris*, accounting for 62% of cumulative model variation. At FRI, no variables were entered.

A similar number of variables produced significant effects in analysis of Japanese climbing fern cover class levels. The first entries into the cover class model were an intercept (value=9.2), followed by importance values for *Carya aquatica,* accounting for a cumulative 17% of model variation. A significant increase in model strength came with additional of the principal components analysis factor FLOOD1, raising model accuracy to 38%. Six tree species: *Catalpa bignoniodes, Platanus occidentalis, Nyssa ogeechee, and Quercus laevis* were accepted in the regression analysis, accounting for a total cumulative model variation of 75%. When the cover class analysis was conducted for each respective site, the only variable entered into the model was FLOOD1 at the FRI site. No variables were entered for BRSF, or the Miller site.

Neural Network Analysis

The neural network analysis selected thirty plot variables and tree importance values for input into the model. Variables positively related to Japanese climbing fern presence are indicated with a (+), while variables positively related to absence are indicted with (-). In order of rank in the model, they include: understory percent cover (-), position (-), *Pinus palustris* importance (-), *Quercus nigra* importance (+), *Ligustrum sinense* importance (+), soil iron (-), percent exposed soil (-), basal area (+), *Quercus falcata* importance (-), *Nyssa sylvatica* importance (-), percent slope (+), soil phosphorus (+), percent canopy cover (+), site, soil aluminum (-), soil calcium (+), hydrology class (+), soil magnesium, *Bumelia lycoides* importance (-), *Quercus stellata* importance (-), *Liquidambar styraciflua* importance (+), soil pH (+), *Quercus laevis* importance (-), *Taxodium distichum* importance (+), soil sodium (+), soil potassium (+), midstory percent cover (-), months flooded (2003) (+), relief (-), and soil organic matter percent (+). Utilizing these variables, the neural network correctly classified Japanese climbing fern presence for 22 of 29 plots, or 76% of the time; Japanese climbing fern absence was correctly classified for 38 of 46 plots, or 83% of the time.

Discussion

In field observations, some trends in the presence and percent cover of Japanese climbing fern are observable. Identification and quantification of the specific causal agents of these trends is experimentally challenging, but some general relationships are discernible from the results of this study. Some aspects of study design, primarily a limited number of plots study-wide; and the macroplot sampling design restrict application of most results beyond the study sites themselves. The following discussion begins with relationships within three main variable categories: site overview, plot environmental variables, and tree species. The final two sections, multiple logistic regression, and neural network analysis, focus on relationships across variable categories, seeking the most valid or predictive combination of variables.

Twenty plot environmental variables and eight tree species demonstrated significant variance in mean values related to the presence or absence of Japanese climbing fern (Tables: A-1, A-2, A-3, A-4) in initial analyses. The majority of these significant variables and tree species either affect or indicate aspects of site hydrology (and associated soil chemistry) and photosynthetic opportunity (e.g., hydrology class, percent flooded, photosynthetically active radiation). This reflects general fact that in Florida, hydrology, pH and fire exert overwhelming influence over site vegetation, minimizing effects of other factors; including soil nutrient supply (Brown, Stone, and Carlisle 1990). The sensitivity of statistical tests used in each analysis should be considered in interpretation of all results, particularly in consideration of forested sites outside the local region, or with different natural community types represented.

Site Overview

Percent cover of Japanese climbing fern varied significantly among sites (Table 2-1). The general grouping of natural community types by site affects a broad range of variables, including Japanese climbing fern occurrence. The primarily hydric Florida River Island (FRI) site had the highest site-level and plot-level percent cover of Japanese climbing fern. Low areas and swales on this site flood annually with the elevation of water levels in the winter and spring like the majority of floodplain and bottomland hardwood forests along the Apalachicola River. Under these conditions, soil saturation is the primary limiting factor affecting climbing fern establishment and survival. Higher elevation areas in this forest type can become heavily infested with Japanese climbing fern as a result of their generally mesic to hydric nature, and extensive flood-based dispersal of spores and on site via the annual floods (Figure 2-5). In addition to site characteristics, regional invasion history may also contribute to higher presence and percent cover of Japanese climbing fern for sites in the Apalachicola River basin (BRSF and Miller site), where the plant has been established for several decades. The xeric to mesic Blackwater River State Forest had the lowest site-level percent cover of Japanese climbing fern (Table 2-1). This reduced site-level occurrence is primarily determined by the spatially limited nature of sites with appropriate hydrology (Figure 2-3), although Japanese climbing fern is increasing in occurrence in both longleaf pine (*Pinus palustris*) and slash pine (*Pinus elliottii*) forests on the site. The majority of Japanese climbing fern on the mesic to hydric Miller site occurred in the pine plantation area and ecotonal areas between upland and lower swamp hardwoods on the eastern portion of the site (Figure 2-4, Table 2-2).

The number of plots with Japanese climbing fern present increased by one on both the Blackwater River State Forest and Miller sites from 2002 to 2004. Foresters and land managers throughout Florida have also noted visually obvious increases in Japanese climbing fern cover and distribution throughout the north Florida region over the last decade (Ferriter 2001). This trend is likely to continue as local and regional propagule pressure from established plants and populations increases, particularly given the limited nature of environmental restrictions on climbing fern occurrence. Plot-level percent cover of Japanese climbing fern increased by 0.4% from 2002 to 2004 at the Miller site, but decreased by 0.6% at Blackwater River State Forest. Neither change was significant. The decrease in plot percent cover at BRSF may reflect the effect of visually high level of foliar damage caused by the rust fungus *Puccinia lygoddii* in 2004, coupled with a small number of plots with hydrology appropriate for Japanese climbing fern. The increase in plot-level occurrence while also not significant, does document the continued expansion of this species on infested sites, either through natural dispersal, or possibly through human-vectored spore dispersal as a result of repeated entry into the plots by researchers.

Discriminant Analysis

Plot environmental variables

Stepwise discriminant analyses of plot variables (alpha=0.10) furthered support for the role of general site hydrology in determining occurrence of Japanese climbing fern. Specifically, soil calcium, and soil iron, plot percent flooded, and plot understory percent cover discriminated between Japanese climbing fern presence and absence at the study level, maintaining support for the importance of site mesic to hydric hydrology in climbing fern establishment. Clawson, Lockaby and Rummer (2001) found soil calcium to increase as flooding levels increased on soils ranging from somewhat poorly drained to poorly drained within a floodplain forest. In addition to hydrologic variables, site-level analysis identified the tree evenness index as a significant indicator at Blackwater River State Forest, probably due to the naturally low tree evenness in the xeric upland longleaf pine community dominating this site (Figure 2-3). As a result, the significance of evenenss in this analysis probably reflects site hydrology and natural community type, rather than indicating a strong relationship between climbing fern occurrence and eveneness. This is further supported by the high occurrence of Japanese climbing fern on the *Pinus elliottii* plots at Miller, which also had low evenness values. Tree species richness was significant at Florida River Island, primarily due to the low richness measured on swale plots (Figure 2-4) which remain inundated for a large part of each year, thereby reducing vegetative establishment opportunity for many species, including Japanese climbing fern.

Discriminant analysis of plots with Japanese climbing fern for relationships with varying cover class levels identified three plot variables as discriminators. Basal area was significant at the study level, driven by the polynomial relationship to Japanese climbing fern cover classes at Florida River Island, where basal area decreases significantly as Japanese climbing fern percent cover exceeds 25%. Plot percent flooded displayed a negative discriminatory relationship with Japanese climbing fern cover, indicating that while adequate moisture is necessary for establishment, excessive moisture (and shortened growing season) will limit percent cover.

Plot tree species

Tree species showing significant relationships to Japanese climbing fern presence (Table A-1) are primarily classified as facultative wetland or obligate wetland species according to the vegetative index used for wetland delineation in Florida (Table 2-3 and Table A-4) (FLDEP 2006). Facultative upland species and facultative species were significantly less important than facultative wetland and obligate wetland species on sites with Japanese climbing fern present, indicating an association with mesic to hydric site conditions. In comparison, no significant difference was measured between vegetative indices on plots with Japanese climbing fern absent, indicating that absence is affected by more than vegetative index of dominant species (e.g., establishment opportunity).

Stepwise discriminant analysis of tree species importance values identified *Quercus nigra, Liquidambar styraciflua,* and *Taxodium distichum*, as significant discriminators of Japanese climbing fern presence. Conversely, *Nyssa sylvatica* was a significant discriminator of Japanese climbing fern absence at the study level. Site-level analyses also identified the invasive plant *Ligustrum sinense* as a discriminator of Japanese climbing fern presence at Blackwater River State Forest. The relationship shown between these species indicates two key factors which are easily recognizeable in the field: the hydrologic function of the extensive natural stream drainageways on the site provide a moisture regime allowing for establishment of Japanese climbing fern and *L. sinense* across the site landscape; and movement of water through the system of drainageways on the site has probably played an important role in vectoring propagules of both species. The natural function and management of these sites are both complicated by the co-occurring populations of these species, as well as an additional non-native invasive plant, *Lonicera japonica*. Removal of the often dense canopy and/or midstory of *L. sinense* has resulted in rapid expansion of Japanese climbing fern in multiple cases. In addition to the documented relationship at Blackwater River State Forest, the pine plantation plots at the Miller site also had co-occuring infestations of Japanese climbing fern and *L. sinense*, though plants were too small to census with plot trees

Pinus elliottii is a significant discriminator for the presence of Japanese climbing fern on the Miller site, as a result of the *P. elliottii* plantation covering the western portion of the study area (Figure 2-4). This site bias does reflect a significant regional relationship between *P. elliottii* plantations and Japanese climbing fern. High levels of Japanese climbing fern invasion are clearly visible throughout mesic pine plantations in the Apalachicola River basin in Florida. One study documented a 22% Japanese climbing fern occurrence level in North Florida *P. elliottii* plantations, a forest type covering 5.1 million acres in the State (Van Loan 2006). Silvicultural management practices on these sites may be contributing to Japanese climbing fern dispersal and site occupancy levels as a result of canopy removal, soil disturbance, and equipment-based spore dispersal. Significant increases have been seen in fern coverage and density on flatwoods and mesic plantation forest types following timber harvest, pinestraw harvest, and prescribed burning in particular.

Discriminant analysis of plots with Japanese climbing fern for relationships with varying cover class levels identified four tree species as discriminators. Increasing importance of *Carpinus caroliniana* was significantly related to increasing occurrence of Japanese climbing fern. A common associate of *Q. nigra*, *Carpinus caroliniana* is considered to demonstrate best growth and development on rich wet-mesic sites (Burns and Honkala 1965), further supporting the observations of the importance of general site hydrology in affecting Japanese climbing fern establishment and spread on infested sites.

Regression Analysis

The multiple regression analysis further supported the importance of mesic to hydric conditions for Japanese climbing fern establishment and spread on a site. The SOIL1 factor (Tables 2-9 and 2-10), reflecting combined associations between soil phosphorus, soil calcium, and pH, varies in association with site hydrology, particularly flooding. Grouping the results of the discriminant and multiple regression analyses reveals five soil variables (calcium, iron, potassium, phosphorus, and pH) associated with presence or absence of Japanese climbing fern, and one soil variable (aluminum) associated with cover class on infested sites. Each of the soil variables associated with the presence or absence of Japanese climbing fern varies in response to multiple factors of parent material, hydrology, moisture source, and habitat inputs. Taken as a group, calcium, phosphorus, pH, and potassium levels in this study were significantly higher on plots with Japanese climbing fern present, typically the mesic to hydric sites, as opposed to absent. In a simplified comparison, iron and aluminum in this study were significantly lower overall on plots with Japanese climbing fern present; however, analysis of fern presence or absence at the site level provided an expanded perspective.

For many soil variables, a separation existed between values measured at FRI versus the other two sites. Calcium, phosphorus, and pH were all significantly higher at FRI compared with BRSF and Miller, while iron was significantly lower at FRI. Iron availability in solution is influenced by soil pH, aeration, and reactions with organic matter. In particular, iron decreases as pH increases, reaching a minimum at pH 7.4 to 8.5. Additionally, poor soil aeration (reduced oxygen levels) caused by flooding can prompt oxidation and/or reduction of iron when influenced by other conditions (Dixon and Schulze 2002). The saturation of soils on FRI reduces soil iron, increasing its solubility, and allowing its movement out of the surface soil layer (Dixon and Schulze 2002). This effect is different from the brief, infrequent periods of saturation in the upland drainageway at BRSF, which produced higher iron concentrations, probably due to moisture-induced oxidation (Dixon and Schulze 2002). However, the significantly higher site-level occurrence of Japanese climbing fern at FRI is probably linked more to the general site hydrology which prompts the soil variable trends, rather than being associated with the individual soil variables themselves.

The significance of soil aluminum is distinguishing between Japanese climbing fern cover classes on infested plots was an artifact of two situations. Aluminum values on the infested BRSF upland drainageway plots and the mesic Miller plots were significantly higher than FRI values, reflecting the effect of lower pH and extreme weathering in the more sandy soils seen at these sites. Low pH values have been shown to prompt a peak in available soil aluminum as seen in sites which repeatedly flood, and then drain (Darke and Walbridge 2000). The addition of the higher values from the Miller and BRSF plots creates the polynomial relationship observed between Japanese climbing fern cover class and soil aluminum on infested plots.

Aspects of site hydrology heavily influence fluctuation in soil conditions and components. Aluminum and iron are known to be important in controlling the retention of dissolved inorganic phosphorus in wetland soils, providing further support for positive relationships between those three variables (Darke and Walbridge 2000). Darke and Walbridge (2000), and Clawson et al. (2000) recorded increasing levels of soil calcium in soils from a gradient of sites ranging from upland to floodplain swale, explaining the trends in calcium observed, particularly at the site level on Miller and BRSF, both sites including a mixture of pine dominated habitat and hardwood dominated habitat. Unlike FRI which is dominated by hardwoods and slightly acidic to neutral soils. Soil pH varied significantly by site due primarily to hydrology and natural community; with higher pH values (5.5 to 7.2) on the hydric FRI hardwood floodplain forest site, and significantly lower pH values (4.6 to 5.9) on mesic Miller site, followed by the xeric BRSF sandhill site (4.6 to 5.0), a trend seen in these habitat types statewide (Londo, Kusha, and Carter 2006, Platt and Schwartz 1990).

As in the discriminant analysis, the effects of *Quercus nigra* and *Carya aquatica* influenced Japanese climbing fern occurrence. These two species, together with *Ligustrum sinense* may be used as general indicators of site conditions appropriate for Japanese climbing fern occurrence in northwest Florida. However, primary application of this result should be restricted to similar natural community types on the same managed areas as utilized in the study. Several years of unquantified observations support the finding that Japanese climbing fern presence may be strongly correlated with the presence of *L. sinense* on a landscape-scale in North Florida. This tendency toward co-location may also extend into Georgia, where *L. sinense* is widely established in upland drainageways, bottomland hardwood and floodplain forests, sites with higher levels of Japanese climbing fern in North Florida.

Neural Network Analysis

Artificial neural networks are attempts to predict complex, non-linear relationships based on a loose analog with living nervous systems. These networks have been used to classify samples according to discrete classes (Mehrotra, Mohan and Ranka 2000), in this case the presence or absence of Japanese climbing fern. Though the network performed relatively well in classification of plots with climbing fern present and absent, the number of variables included (30) indicates overall model weakness in a similar sense to loss of degrees of freedom in regression analysis. However, the predictive performance does allow for some interpretation if primarily for the managed areas on which study sites were located.

While, the model primarily selected variables which had demonstrated significant relationships with Japanese climbing fern presence or absence in previous analyses, it selected some new variables, including position, relief, midstory percent cover, and upland tree species such as *Quercus stellata* and *Quercus falcata*. The addition of these factors in the model network may indicate non-linear relationships that standard techniques cannot correctly model due to non-collinearity or other statistical assumptions in these models. It is also likely that some factors are only useful for this particular dataset, due the limited sample size.

However, all of the data available were used in the fitting of this model, leaving none for model validation. The basic model output does provide support for the initial study hypothesis that a combination of site variables may be identified that affect the presence or absence of Japanese climbing fern on a site. However, the model itself serves only as a hypothesis without independent validation. If future research is planned on classification of sites vulnerable to Japanese climbing fern invasion, a recommendation for study design is to use a larger number of plots on which a smaller number of variables measured may enable use of more robust analyses, provide adequate data for model validation, and reduce some amount of correlation between variables. Data gathered in any future site classification studies may be useful in validation of this model, or construction of others.

Conclusions

The reproductive strategy of Japanese climbing fern plays a significant role in the establishment and spread of this species. Baker's Rule states that species that are capable of uniparental reproduction are more likely to be successful invaders, (Richardson 2004). The results of Foxcroft, Richardson, Rouget and MacFayden (2004) suggest that propagule pressure can act as a fundamental driver of invasions, significantly reducing the importance of environmental limitations (Volin et al. 2004). This situation may at least partially explain the lack of strong results from this study.

However, some relationships between site characteristics and Japanese climbing fern invasion do exist, the most influential of which is general site hydrology. Hydrology has a major role in determination of natural communities and subsequent site environment and vegetation. In particular, flooding extent and duration have several impacts on vegetative communities, affecting growing season duration and establishment opportunity, and soil biogeochemical processes.

A similar assessment examined the correlation between the distribution of Old World climbing fern and abiotic factors in two forested wetlands in South Florida (Volin et al. 2004). In that study, Old World climbing fern occurred on 32% (n=43) of the sampling units, a similar ratio to this study in which Japanese climbing fern occurred on 39% (n=29) of the sampling units (plots). Differences in the sampling design and availability of existing distribution data permitted use of ordination analyses not appropriate for this data set (Volin et al. 2004). However, general conclusions were similar to those in this study. Similar to Japanese climbing fern in North Florida, presence of Old World climbing fern was found to be significantly correlated with site hydrology, specifically a wet but not permanently inundated condition.

Future studies of Japanese climbing fern invasion establishment ecology should be focused in the northern extent of it's range in the southeastern United States. If propagule pressure is the primary determinant of the invasive potential of this species, the majority of mesic and many hydric sites in Florida may already be vulnerable to Japanese climbing fern invasion. Rather than focus heavily on site vulnerability, a critical next step in study of the ecology of this species is to quantify the effects of invasion on habitat quality and function, and native species diversity. The answers to these questions will be key in understanding the impact of Japanese climbing fern invasions and accurately determining the true priority of aggressive prevention and control actions.

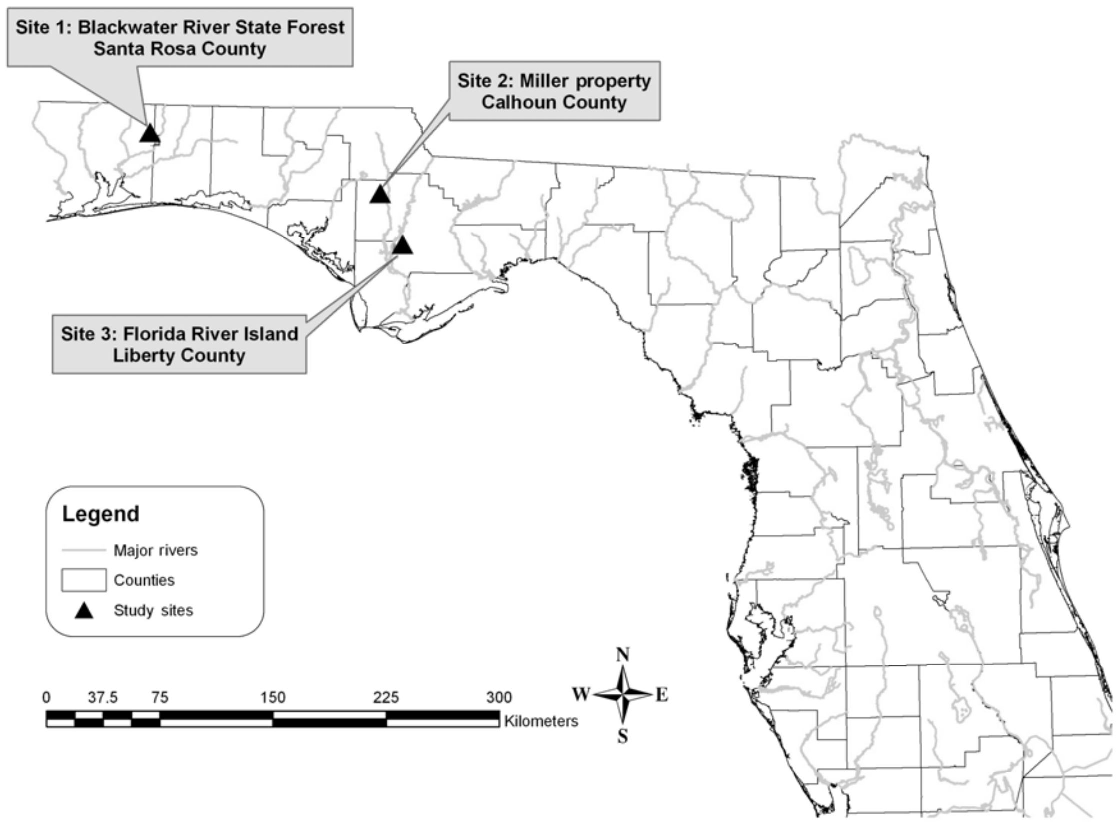

Figure 2-1. Location of study sites used in 2002 Japanese climbing fern site establishment assessment, Florida, USA.

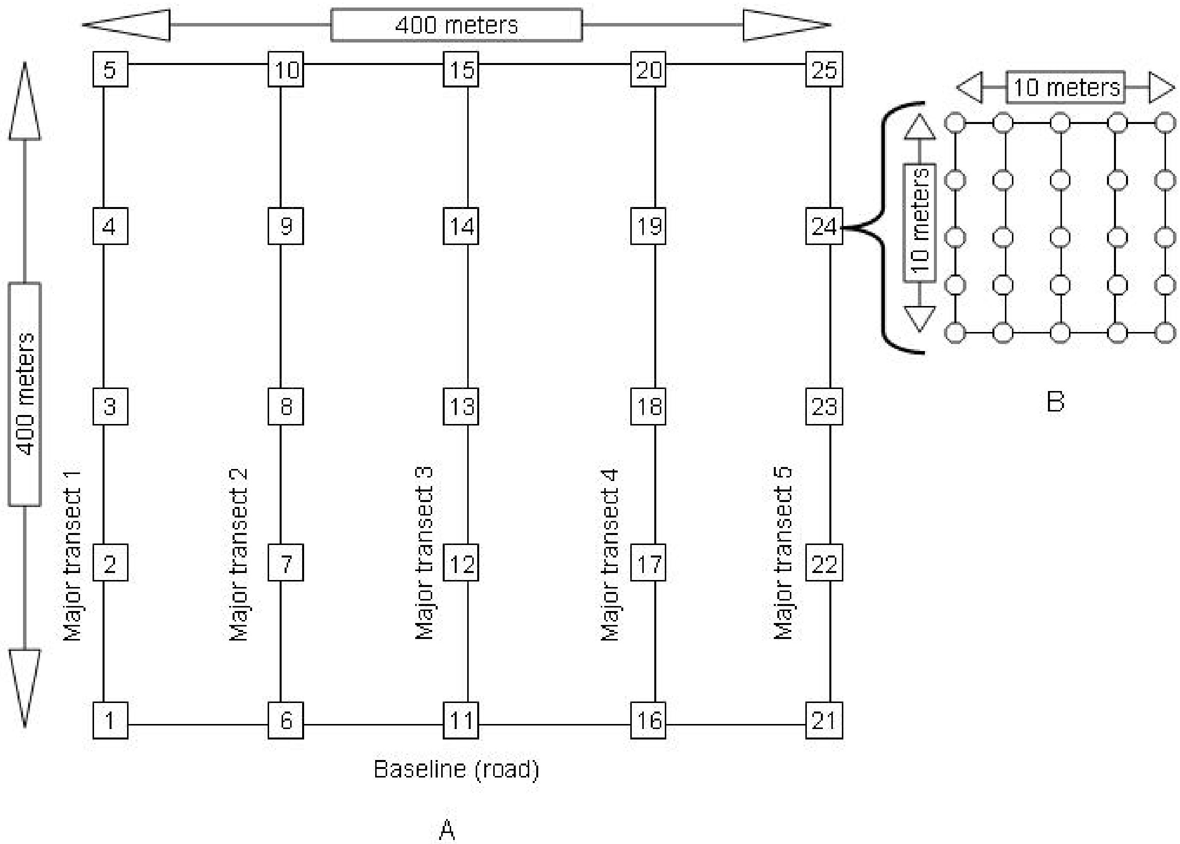

Figure 2-2. Sampling design for assessment of Japanese climbing fern distribution on three forest sites in North Florida. A) Layout of major transect lines and intensive sampling plots. B) Layout of sampling points within each intensive sampling plot

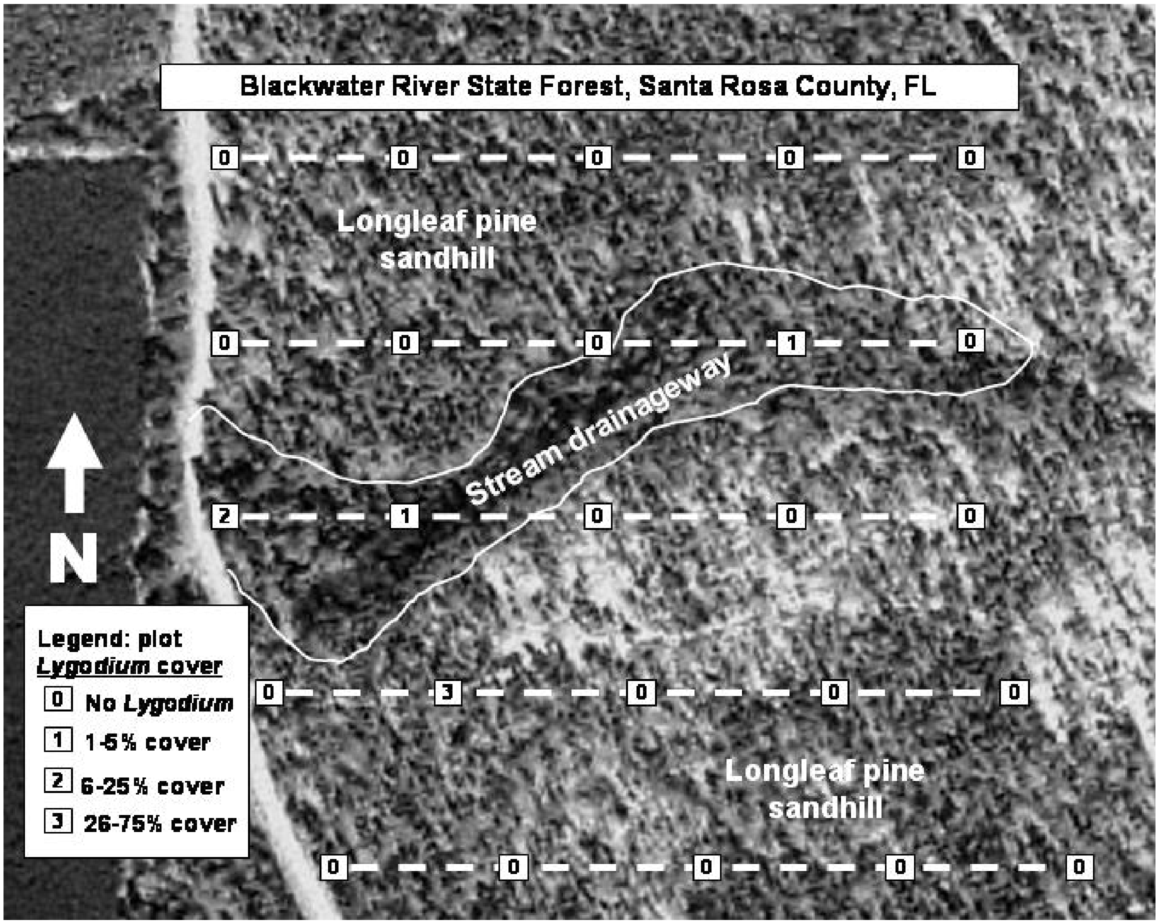

Figure 2-3. Plot layout indicating 2004 Japanese climbing fern cover classes and natural community types, over 2000 aerial imagery, Blackwater River State Forest (BRSF), Santa Rosa County, Florida.

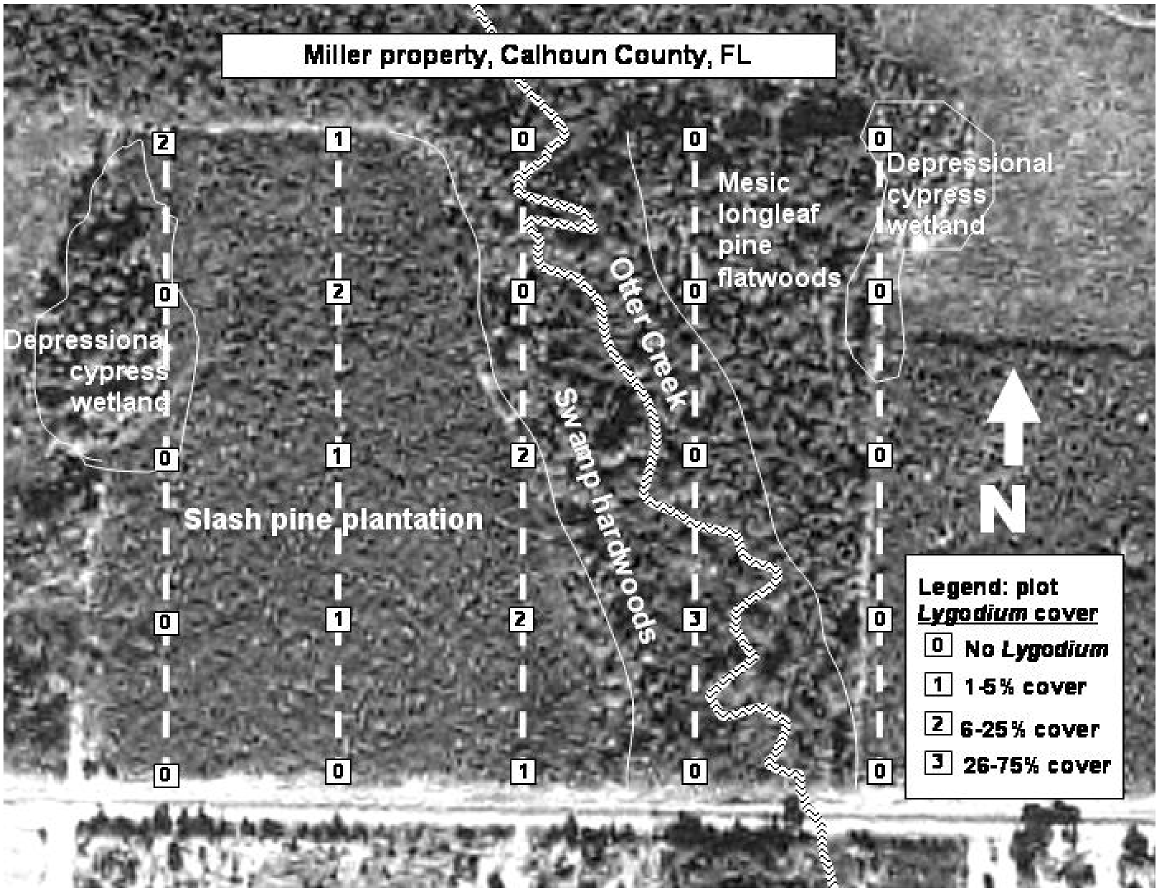

Figure 2-4. Plot layout indicating 2004 Japanese climbing fern cover classes and natural community types, over 2000 aerial imagery, Miller property (Miller), Calhoun County, Florida.

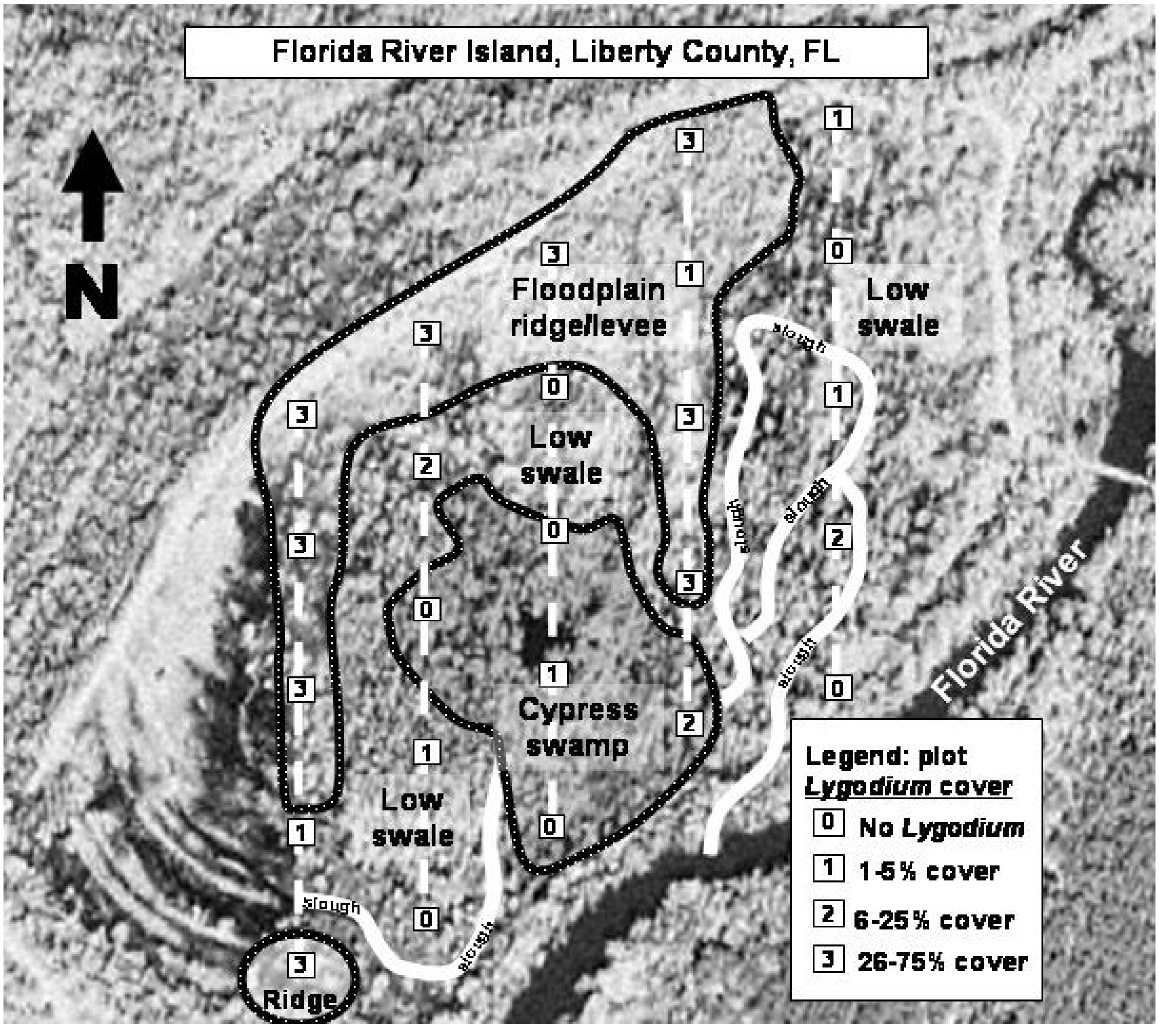

Figure 2-5. Plot layout indicating 2002 Japanese climbing fern cover classes and natural community types, over 2004 aerial imagery, Florida River Island (FRI), Liberty County, Florida.

Table 2-1. Japanese climbing fern (*Lygodium japonicum*) occurrence on three North Florida forest sites, 2002 and 2004.

	Site = BRSF		Site = Miller		Site = FRI	
	2002	2004	2002	2004	2002	2004
Average site % *Lygodium* cover (major transects).	1%	NA[a]	3%	NA	14%	NA
Total number of plots[b] with *Lygodium* present.	3	4	8	9	18	NA
Percent of plots with *Lygodium* present	12%	16%	32%	36%	72%	NA
Average plot % *Lygodium* cover.	3%	2%	5%	6%	14%	NA
Change in *Lygodium* plot % cover 2002-2004.		-0.6%		+0.4%		NA

[a]NA indicates measurement not available.
[b]Twenty-five plots per site.

Table 2-2. Japanese climbing fern (*Lygodium japonicum*) occurrence across eight natural community types in three North Florida forest sites, 2004. Twenty-five plots assessed per site.

	Site = BRSF		Site = Miller		Site = FRI		Site = All	
Natural community	Plot ratio[a]	Plot %[a]	Plot ratio	Plot %	Plot ratio[a]	Plot %[a]	Plot ratio	Plot %
Xeric pine uplands	0/19	0%						
Xeric plot total							0/19	0%
Mesic pine plantation			8/11	73%				
Mesic pine flatwoods			0/5	0%				
Mesic pine/hardwood	4/6	67%						
Mesic plot total							12/22	55%
Hydric floodplain ridge					10/10	100%		
Hydric floodplain swale					6/10	60%		
Hydric cypress wetland			0/4	0%	2/5	40%		
Hydric hardwood swamp			1/5	20%				
Hydric plot total							16/34	47%

[a]Plot ratio and plot % indicate the number of plots with *Lygodium* present to total number of plots of the community type.

Table 2-3. Comparison of tree importance values (IV) by species vegetative index and presence or absence of Japanese climbing fern (*Lygodium japonicum*).

Vegetative index[a]	Number of species	*Lygodium* status	Mean IV	Std. error IV	T-values[b] (present vs. absent)	ANOVA statistic[c]
Facultative upland	5	Present	7	3	4.2 (P<0.0001)*	5.9 (P=0.0005)*
Facultative	13	Present	3	3	-1.3 (P=0.20)	
Facultative wetland	12	Present	10	4	-1.67 (P=0.1)	
Obligate wetland	13	Present	8	4	-2.56 (P=0.01)*	
Facultative upland	5	Absent	25	18		44.9 (P<0.0001)*
Facultative	13	Absent	2	1		
Facultative wetland	12	Absent	7	3		
Obligate wetland	13	Absent	5	2		

[a] Vegetative index of tree species hydrologic associations.
[b] Test statistic and p-values for comparison of mean importance values by Lygodium status (present versus absent) for each vegetative index, Two sample t-test, alpha = 0.05, SAS Version 9.1. Asterisk* indicates significant value.
[c] Test statistic and p-values for comparison of tree importance values between vegetative indices for each Lygodium status (present and absent), one-way analysis of variance, alpha=0.05, SAS Version 9.1. Asterisk* indicates significant value.

Table 2-4. Relationships between Japanese climbing fern (*Lygodium japonicum*) percent cover levels and significant plot variables and tree importance values in three North Florida forest sites.

Variable	*Lygodium* cover class[a]	Number of plots	Mean	Std. error	Kruskal-Wallis values[b]	Correlation coefficients[c]	Correlation strength[d]
Basal area (m^2 ha^{-1})	1	10	20	6	9.62 (P=0.01)*	-0.39 (P=0.04)*	Weak
	2	9	30	2			
	3	10	11	1			
Soil Al (mg kg^{-1})	1	10	178	27	6.62 (P=0.04)*		
	2	9	249	44			
	3	10	154	16			
Carpinus caroliniana	1	10	2	0	6.12 (P=0.05)*	0.38 (P=0.04)*	Weak
	2	9	2	0			
	3	10	14	6			
Percent flooded	1	10	45	15		-0.42 (P=0.03)*	Weak
	2	9	29	14			
	3	10	8	3			
Photosynthetically active radiation (µmol m^{-2} s^{-1})	1	10	92	29		0.40 (P=0.03)*	Weak
	2	9	107	31			
	3	10	199	93			
Quercus nigra	1	10	10	3		0.39 (P=0.04)**	Weak
	2	9	17	6			
	3	10	28	7			

[a]Japanese climbing fern cover class levels: 1=>0 to 5%, 2= 5.1 to 25%, 3= 25.1 to 75%.

[b] Asterisk* indicates significant value - Kruskal-Wallis analysis of variance test and p-values alpha = 0.05.

[c] Asterisk* indicates significant value, Pearson's correlation coefficient and p-values, alpha=0.05, SAS Version 9.1.

[d] Correlation strength: weak=0.2-0.49, moderate=0.5-0.79, strong=0.8-1.0

Table 2-5. Plot environmental variables that discriminate between Japanese climbing fern (*Lygodium japonicum)* presence or absence in three North Florida forest sites.

	All plots[b]			Site = BRSF[b]			Site = Miller[b]			Site = FRI[b]		
Variable[a]	R^2	F-value	P>F	R^2	F-value	P>F	R^2	F-value	P>F	R^2	F-value	P>F
Soil Ca (natural log)	0.31	32.72	P<0.0001	0.28	7.25	P=0.01	0.16	4.21	P=0.05			
Percent flooded	0.05	2.47	P=0.07	0.23	5.93	P=0.02				0.13	3.30	P=0.08
Soil Fe (inverse)	0.06	4.72	P=0.03				0.35	12.48	P=0.002			
Understory (% cover)	0.06	4.48	P=0.04									
Tree evenness index				0.18	4.77	P=0.04						
Hydrology class				0.17	4.21	P=0.05						
Months flooded				0.45	15.34	P=0.001						
Species richness										0.20	5.65	P=0.03
Soil K (natural log)										0.14	3.40	P=0.08

[a] Data transformation indicated in parentheses if used.
[b]Test statistics from stepwise discriminant analysis of covariance of plot environmental variables, SAS Version 9.1. Variables listed are significant (alpha=0.15) at either the study-level or site-level as indicated.

Table 2-6. Plot environmental variables that discriminate between Japanese climbing fern (*Lygodium japonicum)* percent cover class levels in three North Florida forest sites.

	All plots[a]			Site = BRSF[b]	Site = Miller[a]			Site = FRI[a]		
Variable	R^2	F-value	P>F		R^2	F-value	P>F	R^2	F-value	P>F
Basal area	0.20	3.23	0.05					0.24	2.26	0.11
Soil Al	0.19	2.92	0.07		0.82	11.36	0.01	0.25	2.52	0.14
Percent flooded	0.18	2.61	0.09		0.62	3.31	0.14			

[a] Test statistics from stepwise discriminant analysis of covariance of plot environmental variables, SAS Version 9.1. Variables listed are significant (alpha=0.15) at either the study-level or site-level as indicated.
[b] Sample size for site=BRSF too small to conduct analysis.

Table 2-7. Tree species importance values that discriminate between presence and absence of Japanese climbing fern (*Lygodium japonicum*) presence or absence in three North Florida forest sites.

	All plots[a]			Site = BRSF[a]			Site = Miller[a]			Site = FRI[a]		
Variable	Partial R^2	F value	P>F	Partial R^2	F value	P>F	Partial R^2	F value	P>F	Partial R^2	F value	P>F
Pinus palustris	0.21	18.9	<0.0001									
Quercus nigra	0.08	6.6	<0.0001	0.94	3.7	<0.0001						
Liquidambar styraciflua	0.10	8.3	0.01									
Taxodium distichum	0.07	5.6	0.02									
Quercus hemisphaerica	0.05	3.3	0.07									
Lindera benzoin	0.05	3.3	0.07									
Nyssa sylvatica	0.05	3.3	0.07									
Ligustrum sinense				0.91	2.2	<0.0001						
Pinus elliottii							0.23	6.9	0.02			
Acer rubrum										0.01	3.5	0.08
Nyssa aquatica										0.21	5.9	0.02

[a]Test statistics, stepwise discriminant analysis of covariance of tree importance values. Species listed are significant (alpha=0.10).

Table 2-8. Tree species importance values that discriminate between Japanese climbing fern (*Lygodium japonicum)* percent cover class levels on plots with fern present in three North Florida forest sites.

	All plots[a]			Site = BRSF[b]	Site = Miller[b]			Site = FRI[a]		
Variable	R^2	F-value	P>F		R^2	F-value	P>F	R^2	F-value	P>F
Carpinus caroliniana	0.18	2.9	0.07							
Quercus nigra								0.29	3.07	0.08
Carya aquatica								0.32	3.02	0.08

[a] Test statistics from stepwise discriminant analysis of covariance of plot environmental variables. Species listed are significant (alpha=0.10).
[b] Sample size for BRSF too small to conduct analysis, no species selected for Miller site..

Table 2-9. Effects of significant plot environmental and tree importance values in multiple logistic regression for relationship to presence or absence of Japanese climbing fern (*Lygodium japonicum*) in three North Florida forest sites.

Area	Effect (in order of entry)[a]	Estimate	R-squared (cumulative)	Chi square score	P>Chi square
All plots	Intercept	0.40			
	SOIL1	1.79	0.40	24.0	P<0.0001
	Ligustrum sinense	0.29	0.46	11.4	P=0.0008
	Quercus nigra	-0.09	0.53	5.7	P=0.02
	Lindera benzoin	-0.17	0.59	9.4	P=0.002
	Acer rubrum	0.03	0.62	4.4	P=0.04
	Quercus laevis	0.01	0.65	3.9	P=0.05
	Nyssa aquatica	0.75	0.68	4.5	P=0.03
BRSF	Intercept	-1.46			
	TREE	1.80	0.49	7.85	P=0.005
Miller	Intercept	-6.39			
	SOIL1	-1.88	0.42	7.93	P=0.005
	Pinus palustris	0.02	0.64	4.99	P=0.03
FRI	No variables entered				

[a] Significant effects in model (SAS Version 9.1).. Principal components analysis factors: SOIL1= soil P, soil Ca, and soil pH; TREE=tree species richness, evenness, and diversity. Model validity is questionable due to maximum likelihood estimate.

Table 2-10. Effects of significant plot environmental and tree importance values in multiple logistic regression for relationship to percent cover class levels of Japanese climbing fern (*Lygodium japonicum*) in three North Florida forest sites.

Area	Effect (in order of entry)[a]	Estimate	R-squared (cumulative)[b]	Chi square score	P>Chi square
All plots	Intercept	9.2			
	Carya aquatica	3.7	0.17	4.3	P=0.04
	FLOOD1	-1.1	0.38	6.6	P=0.01
	Catalpa bignoniodes	-0.1	0.46	5.2	P=0.02
	Platanus occidentalis	-0.1	0.52	4.1	P=0.04
	Nyssa ogeechee	-1.3	0.59	5.5	P=0.02
	Quercus laevis	0.2	0.75	9.0	P=0.003
BRSF	No variables entered				
Miller	No variables entered				
FRI	Intercept	3.21			
	FLOOD1	-1.37	0.50	8.1	P=0.005

[a] Significant effects in model (SAS Version 9.1). Principal components analysis factor: FLOOD1=hydrology class, percent flooded, and months flooded. Model validity is questionable due to maximum likelihood estimate.

CHAPTER 3
HERBICIDE EFFICACY FOR MANAGEMENT OF JAPANESE CLIMBING FERN IN SOUTHEASTERN FORESTS

Introduction

Japanese climbing fern (*Lygodium japonicum*), is a non-native invasive plant widely established in the southeastern United States. In this range, Japanese climbing fern invades mesic and temporally hydric areas, including: floodplain forests, bottomland hardwood forests, marshes, wetlands, secondary woods, moist pinelands (especially flatwoods), limestone outcroppings, and disturbed areas such as road shoulders and rights-of-way (Clewell 1982, Nauman, 1993, Langeland and Burks 1998). Japanese climbing fern does presently occur on xeric sites as well, but to a more limited extent. Heavily infested sites may sustain populations averaging 60-80% cover over large areas, effectively reduce or eliminate native groundcover and understory vegetation, and smother seedlings of overstory tree species (Horovitz et al. 1998, Lott et al. 2003, Zeller and Leslie 2004). Despite these impacts, large information gaps exist on aspects of the biology and management of this plant. For both public and private land managers, identification of efficient and effective control strategies for Japanese climbing fern has become increasingly important.

Recognition of this plant has increased among public conservation land managers in the southern United States since the mid 1990s, aiding in increased detection and reporting. Recognition of Japanese climbing fern by private land managers has also increased since the late 1990s, but management efforts have not increased at a similar rate due to lack of awareness of the non-native invasive status of the plant, research-based best management practices, and financial incentives or support for management of private-lands infestations. Designation as a noxious weed in Florida has been an important part in the process of raising awareness among private forestland managers and members of the forest products industry (e.g., pinestraw) in Florida, and for increasing the demand for effective management guidelines. Stop sale orders have been issued for shipments of Japanese climbing fern-contaminated pinestraw bales in multiple cases by the FDACS Division of Plant Industry (Van Loan 2006), and awareness of the regulatory requirements is affecting some buyer preferences as well.

Forestry and Invasive Plant Impacts

The impact of species' invasions on sustainable forest management is increasing in scale and recognition. Plant invasions have affected biological diversity, forest health and productivity, water and soil quality, and socieoeconomic value. Plant invasions modify forest ecosystems by affecting fire and hydrological regimes, food webs, and recruitment of dominant tree species (Vitousek, D'Antonio, Loope and Westbrooks 1996, Chornesky, Bartuska, Aplet, O'Britton, Cummings-Carlson, Davis, Eskow, Gordon, Gottschalk, Haack, Hansen, Mack, Rahel, Shannon, and Wainger 2005). In managed forests, establishment of invasive plants is increasingly facilitated by expanding human access, fragmentation, and soil or canopy disturbance often associated with silvicultural practices (With 2002, Chornesky et al. 2005). Some estimates place annual economic impact of forest product loss due to invasive species as high as $2 billion in the United States (Pimental et al. 2000). Impacts of invasive plants on the forestry industry in Florida have been estimated at $38 million per year ($15 million in weed control costs, and $23 million in yield losses) (Lee 2005).

Little is known about the economic impact of Japanese climbing fern invasion. Forest managers in Florida have noted impacts including reduction in annual financial return and harvest lease longevity for pinestraw production, an industry which generates $79 million in revenue for forest landowners in Florida annually (Hodges et al. 2004), and reductions in pine seedling survival during reforestation of heavily infested sites. In particular, mesic private forestlands are moderately to heavily infested with Japanese climbing fern throughout much of the northwest Florida region. A 2002 survey (Barnard and Van Loan 2002) conducted across 280 pine plantations in north and west Florida recorded presence of Japanese climbing fern in 22% of the stands on mesic sites, exceeding occurrence of four other regionally significant non-native invasive plants including: cogon grass (*Imperata cylindrica* (L.) Beauv.), Chinese tallow (*Triadica sebifera* (L.) Small), kudzu (*Pueraria montana* var. *lobata* (Willd.) Maesen & S. Almeida), and air potato (*Dioscorea bulbifera* L.). With approximately 5.1 million acres of mesic pine plantations alone in north Florida (Brown 1995), the regional scale of this invasion generates management problems including: need for coordinated and cost-effective management strategies, legal implications affecting harvest and sale of forest products from infested areas, and high probability of continual reinvasion of treated sites.

Management Techniques

Integrated pest management utilizes the most appropriate combination of treatments including: chemical, manual/mechanical, cultural, and biological control techniques. Mechanical and cultural (i.e., prescribed burning) treatments have generally proven ineffective, often promoting Japanese climbing fern re-growth, and possibly vectoring spores. To date, chemical treatments have been most effective, but little exploratory research has been conducted.

A strategy that crosses property boundaries is required for successful management of most invasive species (Chornesky et al. 2005). The reproductive strategy (i.e. spore-dispersal, self-fertilization) of Japanese climbing fern facilitates rapid spread and establishment in remote areas (Lott 2003), and continual re-invasion if all populations in an area are not addressed jointly. Identification of consistently effective treatment guidelines is an important component in developing a management commitment that crosses property boundaries and is implemented by both public and private land managers. Current knowledge indicates that treatment with herbicides will be a required component of successful management of Japanese climbing fern in the United States.

This study evaluates those herbicides which have received preliminary assessment for Japanese climbing fern control, as well as others commonly used on both forestry and vegetation management. The objective of this study were to: (1) evaluate a range of herbicides to be applied postemergence for control of Japanese climbing fern and (2) evaluate the efficacy of multiple rate combinations of glyphosate and metsulfuron, two herbicides expected to have the greatest efficacy against Japanese climbing fern.

Materials and Methods

Study Area

Field experiments to evaluate the effect of postemergence herbicides applied to Japanese climbing fern were established in 2002 and replicated in 2004. All experiments were conducted in a privately-owned slash pine (*Pinus elliottii* Engelm. var. *elliottii*) plantation in Calhoun County, Florida, USA (30°23’N, 85°9’W). The plantation was established in 1979, managed for pinestraw harvest from 1997-1999, and underwent a third row thinning (removal of every third row of pine trees) in 2000. Japanese climbing fern was first recorded on the site in 1997, but may have been present earlier. By 2002, the stand was heavily infested, with Japanese climbing fern dominating the understory and trellising into the midstory. For both of the 2002 trials, the soil was a Dothan sandy loam, 0-2 percent slope (fine-loamy, kaolinitic, thermic Plinthic Kandiudults) with 0.48% organic matter, and a soil pH of 5.3. The 2004 trial was conducted within the same stand, but on slightly different soil; Robertsdale fine sandy loam (fine-loamy, siliceous, semiactive, thermic Plinthaquic Paleudults) with 1.87% organic matter, and a soil pH of 5.2. The average daily temperature is 12° C in the winter and 33° C in the summer. Freezing temperatures occur on an average of 20 days each winter.

Herbicide Selection

Herbicides were selected based on their weed control spectrum, label-status for forestry applications, performance in initial field assessments by land managers, and commonality with best management practices for treatment of other non-native invasive plants regularly co-occurring with Japanese climbing fern in Florida (Table 3-1). A non-ionic surfactant "Induce" (Helena Chemical) was added at 0.25% v/v according to label recommendations.

In response to land manager concerns and requests, two formulations of glyphosate were evaluated; a "traditional" formulation (Accord ™), and a "generic" (Glyphos™) formulation, representing the many glyphosate products available for purchase since the expiration of the Roundup™ patent. Land manager interest in this comparison was primarily due to the lower per unit cost of the "generic" formulations. Both formulations were evaluated at a low application rate (2.24 kg ai per ha, or 2.5% in solution), and a high application rate (4.48 kg ai per ha, or 5% in solution).

Experimental Design

Two herbicide trials were established in October 2002. The "Multiple Herbicide Trial" included fifteen herbicide treatments and an untreated control (see Table3-2). Treatment plot size was 6.8 m by 3.4 m. The "Glyphosate/Metsulfuron Trial" included tank-mixed combinations of four rates of glyphosate (0, 1.12, 2.24, 4.48 kg ai per ha) and metsulfuron (0, 0.03, 0.06, 0.12 kg ai per ha), and an untreated control, for a total of sixteen treatments. Treatment plot size for this trial was 3.4 m by 3.4 m and were smaller than desired due to constraints within the study site. Treatment plots were established in the thinned rows of the stand. All treatments were applied using a CO_2-pressurized hand-held boom sprayer. Spray volume was calibrated to deliver 225 liters per hectare (20 gallons per acre). Experiments were conducted as randomized complete block designs, and all treatments were replicated four times, for a total of sixty four plots per trial. The

"Multiple Herbicide Trial" was replicated within the study site in 2004 with treatment plots sized 6.8 m by 3.4 m. The "Glyphosate/Metsulfuron" trial was not replicated in 2004 due to lack of adequate area with contiguous Japanese climbing fern coverage in the study site, and the expectation of greater utility of results from the "Multiple Herbicide" trial for land managers.

Treatment and Assessment Schedule

2002 trials. Herbicides were applied on October 23, 2002. Percent cover of live Japanese climbing fern was rated at 1 month after treatment (MAT), 5, 6, 8, and 12 MAT. Upon returning to the study site for the 12 MAT rating, it was discovered that the pine stand had been unexpectedly thinned in late September, 2003 (removal of selected trees from "leave" rows remaining after 2000 thinning, and skidding of logs through all plots). Aboveground Japanese climbing fern foliage was mechanically removed from all plots by the harvest operation (percent cover = 0), but plot corner stakes were adequately intact to re-define original plot boundaries. Following harvest, percent cover of live Japanese climbing fern was rated at 15, 24, 29, 31, 35, and 36 MAT.

2004 trial. The 2002 Multiple Herbicide Trial was repeated within the original study site to remove the potential confounding effects due to logging. Treatment plot size was 3.4 by 6.8 m. Herbicides were applied on November 10, 2004. Percent cover of live Japanese climbing fern was rated at 1, 4, 6, 10, and 12 MAT.

Data Analysis

Percent cover of live (green foliar tissue) Japanese climbing fern was visually determined and recorded for each plot to rate effectiveness of the various treatments. All plots were rated on a scale from 0% (no climbing fern on plot) to 100% (plot completely covered by climbing fern) in 5% increments one day prior to treatment to establish a baseline. Following treatment, plots were rated using the same technique according to the assessment schedules indicated above; all ratings were were conducted by the same observer.

For all plots, percent live Japanese climbing fern cover was converted to percent control for analysis using the following equation:

$$\text{Percent control} = \left(\frac{\text{Percent cover pre-treatment} - \text{Percent cover at MAT}}{\text{Percent cover pre-treatment}} \right) \text{X } 100$$

Use of percent control values accounts for differences in pre-treatment fern cover across treatment plots. The results from each trial were analyzed using the general linear model approach to detect significance of model effects among all treatments. Fisher's least significance difference (LSD) test was used to make pairwise comparisons among treatment means (SAS Institute 2005). For all tests, significance was assessed at the alpha=0.05 level.

A two-sample t-test was performed to evaluate the effect of plot size on trial results. Means from untreated control plots in the 2002 Multiple Herbicide trial and the 2002 Glyphosate/ Metsulfuron trial were compared, with the hypothesis that no difference would exist.

Results and Discussion

The reproductive strategy (i.e. spore-dispersal, self-fertilization) of Japanese climbing fern facilitates rapid spread and establishment in remote areas (Lott et al. 2003), and continual re-invasion if all populations in an area are not addressed jointly. Identification of consistently effective treatment guidelines is an important component in developing a management commitment that crosses property boundaries and is implemented by both public and private land managers. A strategy that crosses property boundaries is required for successful management of most invasive species (Chornesky et al. 2005). Current knowledge indicates that treatment with herbicides will be a required component of successful management of Japanese climbing fern in the United States. The following results verify and expand the current knowledge base on herbicide selection for this purpose.

Phenology of Japanese Climbing Fern

To fully understand the herbicide treatment effects for any species, it is important to consider other factors which may influence plant performance. On all plots in this study, three primary factors affected climbing fern foliar cover: herbicide treatment (varied by treatment), seasonal dieback and growth (uniform across treatments), and mechanical damage from the 2003 timber harvest (uniform across treatments). Japanese climbing fern plants remain evergreen below the frost line in central and south Florida (Ferriter 2001, Valenta et al. 2001, Zeller and Leslie 2004). Above the frost line, winter dieback of Japanese climbing fern fronds occurs to varying extents through the majority of its range. In the spring, the plants will re-sprout from cold-tolerant subterranean rhizomes (new stems observed the first week in March in North Florida), and often utilize freeze-damaged stems as ladders to grow back into the canopy. This seasonal dieback in sub-temperate and temperate climates is one factor which has likely prevented formation of the canopy-covering arboreal blankets seen with Old World climbing fern (*Lygodium microphyllum*) (Zeller and Leslie 2004). However, the increasing distribution of Japanese climbing fern in south Florida may lead to the development of such canopy-smothering growth on infested sites.

Japanese climbing fern seasonal foliar phenology was recorded on the untreated control plots in each trial throughout the study period. At inception of the 2002 and 2004 trials (October), untreated Japanese climbing fern plants had achieved maximum annual foliar coverage, followed by a seasonal frost event (December-February) which reduced plot live foliar cover to 11% on average (min. 5%, max 20%). Initial emergence of new croziers (growing shoots) occurred in mid-March, with measurable new growth of climbing fern present by mid-April. Foliar cover continued to increase steadily through the summer and fall until peaking again in October for both the 2002 and 2004 trials (Figure 3-1). Despite these seasonal fluctuations and the 2003 timber harvest event, Japanese climbing fern cover on the control plots averaged a 3.1% annual increase over the initial percent cover (Table 3-3). However, analysis of variance did not identify this change as significant (alpha = 0.05)

Multiple Herbicide Trial: 2002 and 2004

Average initial climbing fern percent groundcover on the Multiple Herbicide trial plots was 70% (min 30%, max 95%). A significant effect was measured for the interaction of treatment and year between 2002 and 2004, and as a result, the data from each year are presented separately. Discussion of “short-term” treatment effects observed through 12 MAT will focus on the 2004 trial results. This ensures continuity not possible in the 2002 plots due to the interruptive effect of the 2003 timber harvest. Results from the 2002 trials are used for discussion of “long-term” treatment effects observed at 24 and 36 MAT. The plot ratings from 5 and 29 MAT, and 15 MAT are not discussed as they represent effects of seasonal frost damage and timber harvest respectively, rather than herbicide treatment effect. Review of results from this trial and those from Valenta et al. (2001), and Zeller and Leslie (2004) indicate that the best performing (“successful”) treatments typically yield a minimum of 70% control at 12 MAT; a standard which will be utilized in the following discussion.

Short Term Treatment Effects

Short term treatment effects reported herein include effects of treatments applied in 2004, from treatment through twelve months after treatment. At 1 MAT (December 2004), three treatments exhibited a level of damage to Japanese climbing fern foliage that was significantly ($P<0.05$) different than observed in the untreated control plots (Table 3-2). Hexazinone and 2,4-D+dicamba showed the highest initial damage to Japanese climbing fern with 91% control (reduction in live foliar cover), followed by the dicamba treatment with 80% control. This initial response is probably due to “burn-off” of the foliage rather than the damage to underground plant structures necessary to achieve long-term control. Similar effects were observed in early measurements of treatments by Valenta et al. (2001), and Zeller and Leslie (2004). Three other treatments yielded control levels greater than 70%: the "high" rate of glyphosate in both the "traditional" (77%) and "generic" (76%) formulations, and the tank mix of glyphosate+imazapyr (72%). The tank mix of glyphosate+metsulfuron yielded 65% control, while the glyphosate treatments at "low" rate yielded 54% and 29% control, respectively, for the "generic" and "traditional" formulations. All other treatments yielded less than 50% control. The untreated control plots yielded a 32% reduction in foliar cover, which may be explained by early winter decline in foliage density observed following the fall "peak" in foliar cover or as a result of foliar rust damage, and/or damage to fronds originating in other treatment plots and growing across adjacent control plots.

AT 6 MAT (May 2005), treatments separated into three significant ($P<0.05$) groups (Table 3-2). The largest group included eleven treatments exhibiting between 89% and 100% control of Japanese climbing fern. This group included all of the herbicides and herbicide tank mixes in the trial with amino acid inhibition as the mode of action: glyphosate (both rates and formulations), imazapyr, metsulfuron, glyphosate+metsulfuron, glyphosate+imazapyr, imazapyr+metsulfuron, imazapic, and sulfometuron. In a statewide survey of climbing fern treatment regimes used by public land managers, many favored a six-month return interval for herbicide treatments (C. Lockhart, 5/04/2006, personal communication). With such a regime, all treatments in this group may be considered in managing Japanese climbing fern. A second group of treatments, including hexazinone, dicamba, and 2,4-D +dicamba, exhibited approximately 60% control, while triclopyr ester dropped to 25%, indicating higher fern coverage than on the untreated control plots (35%). This control level observed in the untreated plots is primarily explained by lingering effects of winter climbing fern dieback.

At 10 MAT (September 2005), two significant ($P<0.05$) groups of treatments were observable. As found at 6 MAT, the group of treatments exhibiting the highest level of control (69% to 94%) included all of the amino acid inhibitors, with the exception of sulfometuron where control levels were reduced to 0.1%. A second group of treatments including hexazinone, dicamba, 2,4-D+dicamba, sulfometuron and the untreated control plots exhibited between 0.1% and 14% control; while triclopyr ester plots showed a negative control level, indicating that climbing fern cover had increased by 12% from the pre-treatment levels. The untreated control plots still yielded 14% control at this assessment as climbing fern foliar cover continued to increase through the remainder of the growing season (Figure 3-1).

At 12 MAT (November 2005), three significant ($P<0.05$) treatment groups were observable (Table 3-2). The herbicide group exhibiting the highest level of reduction in climbing fern included four treatments yielding greater than 90% control (the "traditional" and "generic" formulations of glyphosate at the high application rate, and tank mixes of glyphosate+imazapyr and glyphosate+metsulfuron); and three treatments exhibiting from 73% to 87% control (the "traditional" and "generic" formulations of glyphosate at the low application rate, imazapyr, and the tank mix of metsulfuron+imazapyr.) At this post treatment date, the effects of metsulfuron and imazapic treatments had diminished to 41% and 27% control, respectively, while hexazinone exhibited only 3 percent control. For five treatments, percent cover increased above pre-treatment levels, yielding a negative percent control value, including: dicamba (8%), sulfometuron (11%), triclopyr ester (12%), and 2, 4-D+dicamba (12%), and the untreated control plots (3%).

Glyphosate comparison. Results from the 2004 Multiple Herbicide trials were consistent with initial expectations, in that the treatments that included glyphosate were among the highest performing treatments. Direct comparison of the "traditional" and "generic" glyphosates at both high and low application rates yielded little difference between the two formulations. At 1 MAT, significant difference ($P<0.05$) was detected between the "traditional" and "generic" formulations at the low application rate, but at all subsequent measurements no difference was detected between the two formulations.

Timber Harvest Effects

The October 2003 timber harvest clearly affected all of the 2002 study plots by physically removing the aboveground fern growth, reducing canopy cover across the stand, increasing the amount of downed woody debris, and possibly re-distributing Japanese climbing fern spores across the study site. The harvest effects appeared to be uniform across all plots based on visual assessment. The plots then underwent a period of post-harvest "recovery" which lasted for approximately one year, and during which climbing fern reclaimed the site understory.. Treatment effects became identifiable again at 24 MAT, and lasted through the final assessment at 36 MAT.

Long Term Treatment Effects

Long term treatment effects reported herein refer to effects measured on both the 2002 and 2004 plots from 12 MAT to 36 MAT. Treatment effects which were beginning to appear in the 2002 plots at 8 MAT, and which also were clear in the 2004 plots at 12 MAT, reappeared in the 2002 plots at 24 MAT (Table 3-2). Analyses revealed many overlapping relationships, but yielded two primary treatment groups. Nine treatments exhibited significantly higher ($P<0.05$) levels of climbing fern control (38% to 70%) than seen in the untreated control plots, including: all treatments that included glyphosate ("traditional" and "generic" formulations, low and high rates and tank mixes), and metsulfuron and imazapyr.

Treatment effects remained similar at 36 MAT (Table 3-2). Six treatments continued to exhibit significantly higher levels of climbing fern control (35% to 41%) than observed in the untreated control plots, including both rates of the "traditional" glyphosate formulation and the low application rate of the “generic” glyphosate formulation, imazapyr, and tank mixes of glyphosate+imazapyr and metsulfuron+imazapyr. Seven herbicide treatments exhibited between 29% control (reduction) and 24% increase in climbing fern cover, not significantly different than the untreated control plots. Climbing fern cover on the untreated plots increased above initial pre-treatment levels by 6.25%.

2002 Glyphosate/Metsulfuron Trial

The initial decision to conduct the Glyphosate/Metsulfuron rate combination trial was made based on performance of these two herbicides in preliminary assessments (Valenta et al. 2001, Zeller and Leslie 2004). Metsulfuron showed potential for limiting damage to non-target plant species during invasive plant management. In addition to the recovery of ecosystem value through retention or re-establishment of native species on sites affected by non-native species invasions, several studies have shown the importance of site occupancy in preventing new or recurring site invasion by non-native species, particularly following a disturbance event such as chemical treatment (Burke and Grime 1996, Davis et al. 2000, Prieur-Richard and Lavorel 2000, D’Antonio and Myerson 2002). Identification of a control strategy which limits loss of non-target species may play a significant role in long term success of any Japanese climbing fern management strategy. Zeller and Leslie (2004) observed glyphosate treatments to yield better long term control (70% to 80% reduction in fern cover at one year after treatment) than metsulfuron treatments, but more non-target damage occurred compared to metsulfuron. In sites where herbicide applications are likely to be repeated within a single year, this finding may indicate use of metsulfuron for retention of desirable non-target species. However, concerns exist among weed management specialists about rapid development of resistance to sulfonylurea herbicides such as metsulfuron (Tranel and Wright 2002), particularly with plants which reproduce as rapidly and prolifically as the climbing ferns (Lott et al. 2003). Treatments that involve tank-mixing glyphosate and metsulfuron hold promise as a technique to reduce both the non-target damage seen with glyphosate, and the possibility of herbicide-driven selection of metsulfuron-resistant climbing fern populations (Diggle et al. 2003, Kudsk and Streibig 2003). This concern prompted the evaluation of multiple rate combinations of glyphosate and metsulfuron in this study.

Average initial climbing fern cover on the Glyphosate/Metsulfuron plots was 66% (min 30%, max 90%). As with the 2002 Multiple Herbicide plots, percent groundcover of live Japanese climbing fern was rated at 1, 5, 6, 9, and 12 MAT. Plots were affected by thinning before 12 MAT, at which time percent cover for all plots was mechanically reduced to zero. In the period following harvest, plots were rated at 15, 24, 29, 31, 35, and 36 MAT.

The plot size for the Glyphosate/Metsulfuron trial was 3.4 m by 3.4 m, 50% smaller than the plot size for the Multiple Herbicide trials. This factor was significant in determining the usefulness of the trial results (data not shown). At nearly all assessment dates, the percent control of Japanese climbing fern on the untreated control plots was not significantly different ($P<0.05$) than the herbicide treatment plots. The untreated control plot data from the Glyphosate/Metsulfuron trial and the 2002 Multiple Herbicide trial were compared to assess the effect of plot size. Significant differences ($P<0.05$) due to plot size were seen at 5, 6, 8, and 24 MAT, indicating that the treatment differences detected in this study represented a Type 1 error rather than actual treatment differences. A recommendation for future research is to limit the minimum herbicide trial plot size to 3.4 m by 6.8 m (200 square feet). Further evaluation of these treatments may be appropriate; however, in the interest of achieving long-term control of Japanese climbing fern a higher priority may be placed on research which evaluates the effect of repeated herbicide applications.

Conclusions and Recommendations for Land Managers

The typical goal of invasive plant management on public conservation lands is, when possible, to eradicate the species from infested sites in such a way as to promote maximal recovery of native plant species following treatment. This may also be the management goal on some private forestlands, but for many others, preservation of native midstory and groundcover species is secondary to production of a profitable and legally saleable forest product (Allen et al. 2005). In the case of the pinestraw industry, the vegetation management goal is typically to remove understory vegetation from a harvest site that may impede the pinestraw harvest process, or serve as a non-straw contaminant in the pinestraw bales (Duryea 1998, Taylor and Foster 2003). These varying goals will impact implementation of the results of this study by land managers in the field.

The data of greatest interest to land managers are the results from the 2004 Multiple Herbicide trial in the 0 to 12 MAT period. Increasingly, a six month return interval is recommended or implemented in climbing fern management (Ferriter 2001, Lockhart 2006). While managers often strive to revisit treatment sites multiple times in a year, many operational invasive plant management programs on public and private lands include a functional minimum return interval of approximately one year between herbicide applications. However, budget and staff limitations can prevent mangers from achieving this minimum, resulting in return intervals which may extend to two years or more.

The long term (>12 MAT) results from this trial illustrate that even when such extended return intervals occur, some treatment effects will remain, to the point of reducing the Japanese climbing fern coverage by 39% on average across the "good" treatments at three years post-treatment (Table 2). While positive in some sense, this extended suppressive effect should not prompt land managers to delay return to a site for management purposes, as the species' spore-based reproductive strategy allows even small infestations to contribute significantly to re-infestation of a site, and spread to other sites. In addition, the vacancies created in areas where herbicide treatments are successful, are most likely to be filled by either Japanese climbing fern or another non-native invasive plant, providing additional incentive for regular follow-up management visits (Davis et al. 2000, D'Antonio and Myerson 2002).

Land managers will consider three main factors when determining which approach to use in treatment of Japanese climbing fern: level of control, duration of control, and per area treatment cost. As indicated earlier, a minimum level of control in successful treatments may be 70% reduction of Japanese climbing fern foliage (Table 3-4). However, in this study four treatments (glyphosate high rate "traditional" and "generic", glyphosate + imazapyr, and glyphosate + metsulfuron) maintained greater than 90% control for up to 12 MAT (Table 3-2). This highest level of control also limits spore production, a factor which is very important in the long term management of this plant. Three treatments from this trial provided control for 6 (sulfometuron) to 10 (metsulfuron and imazapic) MAT. Managers may decide to treat Japanese climbing fern with one of these treatments as a secondary component of herbicide application for other management purposes, or in the case of metsulfuron, if reduced damage to non-target species is a management priority. Sulfometuron has been recommended for herbaceous weed control in hardwood plantations both preplanting and postplanting (Rhodenbaugh and Yeiser 1994, Ezell and Nelson 2001). Bottomland hardwood, floodplain forest, and mixed pine-hardwood stands are both readily invaded by Japanese climbing fern in Florida, and in some cases actively managed for hardwood production. In such sites, sulfometuron can be applied to dormant hardwood seedlings, and over conifer seedlings (Schuler, Robison and Quicke 2004). However, any damaging effects of these compounds on Japanese climbing fern will likely only last 6 months, and require attention to regular follow-up treatments.

Based on level of control achieved and approximate prices in 2006 (Ferrell, Gray and MacDonald 2006), the glyphosate treatments were the most cost-effective, and efficacious at both low and high application rates using either the "traditional" or "generic" formulations. Glyphosate is widely used in many aspects of forestry and invasive plant management, as well as in other vegetation management applications. Valenta et al. (2001) and Zeller and Leslie (2004) also found glyphosate treatments to be among the most effective, achieving greater than 70% control at 0.07, 1.34, and 2.67 kg ai/ha at 315 days after treatment, and 1.12 and 3.36 kg ai/ha at 400 days after treatment, respectively. This wide use facilitates the ready expansion of treatment protocols established for other purposes to include Japanese climbing fern.

Imazapyr is another herbicide widely used in pine plantation management (Lauer et al. 2002) and invasive plant management in the South. Though currently less cost effective than glyphosate, imazapyr is effective against Japanese climbing fern when applied alone and in tank mixes with glyphosate and metsulfuron; residual effects were observable up to 36 MAT. Though effective, selection of imazapyr for Japanese climbing fern management should include consideration of soil activity and associated non-target species damage (Tu, Hurd and Randall 2001). Clark (2002) found no significant difference in native plant cover, richness or diversity on sites infested with Old World climbing fern that were either treated with herbicide or left untreated; illustrating that with heavy infestations, the damaging effects of chemical treatments may be no greater than the effects of heavy infestation.

Regardless of which "successful" treatment is selected, managers must be vigilant in completion of regular follow-up treatments to reduce Japanese climbing fern infestations to control levels acceptable within their management priority framework.

Spore contamination during treatment. When implementing any management program for Japanese climbing fern, spore vectoring on contaminated personnel, clothing, and equipment must be considered. Hutchinson and Langeland (2006) evaluated Old World climbing fern gametophyte (germinated spore) formation from spores collected on clothing and equipment of herbicide applicators after working in three heavily infested sites receiving an initial herbicide treatment, and one site with a very low infestation undergoing a re-treatment. Contamination of clothing and equipment occurred in the initial and re-treatment sites, affecting shirts, pants, sprayers, disposable suits, chainsaws, gloves, machetes and footware. Contamination was significantly greater from the heavily infested initial treatments sites versus the re-treatment sites. Similar results are likely in sites infested with Japanese climbing fern, and applicators working in these sites should utilize the following "good spore hygiene recommendations": spray off equipment and brush off clothing and accessories before leaving site, wash all clothing daily, store disposable suits in plastic bags, limit any vehicle entry into infested areas (Hutchinson and Langeland 2006.)

This study has confirmed and expanded knowledge of the efficacy and treatment duration of several herbicide rates and combinations for use in management of Japanese climbing fern, particularly for forest managers in the Southeast United States. However, additional research may clarify other aspects of the treatment of this species. Targets for future research may include: further assessment of the impacts of herbicide treatments on non-target native species, measurement of effects of repeated treatments, and assessment of mechanical, biological and other treatment techniques for efficacy in an integrated management approach. Finally, a comparison of infestation levels and treatment efficacies in disturbed versus undisturbed sites may improve consideration and prioritization of Japanese climbing fern infestations in sites schedules for silvicultural management practices.

Table 3-1. Herbicide chemical families and mode of action used in a Japanese climbing fern control trial in a North Florida pine plantation, 2002 to 2005.

Active ingredient	Family[a]	Mode of action[a]	Mechanism of action[a,b]	Absorption[a]	Accumulation[a]	Plant death[a]	Soil half life[b]
Imazapic	Imidazolinone	Amino acid inhibitor	ALS inhibitor	Leaves, stems, roots	Underground tissues	several weeks	120 days[c]
Imazapyr	Imidazolinone	Amino acid inhibitor	ALS inhibitor	Leaves, roots	Meristematic regions	1to 2 weeks	25 to 142 days (3 to 24 month)[c]
Sulfometuron	Sulfonylurea	Amino acid inhibitor	ALS inhibitor	Leaves, roots	Meristematic regions	2 to 3 weeks	20 to 28 days
Metsulfuron	Sulfonylurea	Amino acid inhibitor	ALS inhibitor	Leaves, roots	Meristematic regions	1 to 2 weeks	1 to 6 weeks
Metsulfuron + imazapyr	Sulonylurea + imidazolinone	Amino acid inhibitor	ALS inhibitor	Leaves, roots	Meristematic regions	1 to 2 weeks	see Imazapyr
Glyphosate	Glyphosate	Amino acid inhibitor	EPSP inhibitor	Leaves, green stems	Underground tissues, young leaves, meristems	4 to 7 days	47 days
Glyphosate + imazapyr	Glyphosate + imidazolinone	Amino acid inhibitor	EPSP + ALS inibitors	Leaves, stems, roots	Underground tissues, young leaves, meristems	1 to 2 weeks	see Imazapyr
Glyphosate + metsulfuron	Glyphosate + sulfonylurea	Amino acid inhibitor	EPSP + ALS inibitors	Leaves, stems, roots	Underground tissues, young leaves, meristems	1 to 2 weeks	see Glyphosate
Dicamba	Benzoic acid	Growth regulator	Synthetic auxin	Leaves, stems, roots	Growing points	3 to 5 weeks	14 to 28 days
Triclopyr ester	Picolinic acid or carboxylic acid	Growth regulator	Synthetic auxin	Leaves, roots	Growing points	3 to 5 weeks	10 to 46 days

Table 3-1. Continued

Active ingredient	Family[a]	Mode of action[a]	Mechanism of action[a,b]	Absorption[a]	Accumulation[a]	Plant death[a]	Soil half life[b]
2,4-D + dicamba	Phenoxy acetic acid + benzoic acid	Growth regulators	Synthetic auxin	Leaves, roots	Growing points of root and shoot	3 to 5 weeks	1 to 4 weeks
Hexazinone	Triazine	Photosynthetic inhibitor	Binds to D1 protein in photosystem II	Leaves, roots	Leaves		90 days (soil:1 to 6 months, leaf litter: to 3 years)

[a] Ahrens 1994
[b] ALS = Acetolactate synthase, EPSP = 5-endolpyruvyl -shikimate-3- phosphate synthase
[c] Tu et al. 2001

Table 3-2. The effect of post-emergent herbicides on Japanese climbing fern foliar cover in a North Florida slash pine plantation, 2002 and 2004.

Treatment[c]	Rate	1 MAT[b] 2002[d]	1 MAT[b] 2004	6 MAT 2002	6 MAT 2004	8 MAT 2002	10 MAT 2004	12 MAT 2002[e]	12 MAT 2004	24 MAT 2002	36 MAT 2002
	kg ai/ha	% reduction in Japanese climbing fern cover									
Untreated control	-	7 a	32 bc	38 a	35 a	25 a	14 b	-	-3 a	-10 a	-7 ab
Glyphosate "traditional"	2.24	10 a	29 abc	96 d	89 c	91 c	69 c	-	73 d	70 f	38 c
Glyphosate "tradtional"	4.48	7 a	77 gh	94 d	100 c	91 c	91 cd	-	91 d	51 def	41 c
Glyphosate "generic"	2.24	5 a	54 def	94 d	98 c	91 c	89 cd	-	87 d	56 ef	40 c
Glyphosate "generic"	4.48	7 a	76 gh	92 d	97 c	93 c	93 d	-	92 d	38 bcdef	19 bc
Metsulfuron	0.063	9 a	27 ab	98 d	100 c	95 c	74 cd	-	41 c	43 cdef	13 abc
Imazapyr	0.84	10 a	48 cde	100 d	100 c	92 c	90 cd	-	77 d	47 cdef	35 c
Imazapic	0.56	6 a	32 bc	100 d	100 c	100 c	81 cd	-	27 bc	29 abcdef	14 abc
Sulfometuron	0.16	8 a	9 a	73 c	91 c	51 b	0.1 ab	-	-11 a	15 abcde	0.4 abc
Dicamba	3.36	18 ab	80 gh	53 b	57 b	26 a	2 ab	-	-8 a	4 abc	-24 a
Triclopyr ester	1.12	28 bc	40 bcd	60 b	25 a	44 b	-12 a	-	-12 a	11 abcd	5 abc
Hexazinone	3.36	87 d	91 h	52 b	62 b	37 ab	14 b	-	3 ab	-1 ab	4 abc
2,4-D + dicamba	3.36	41 c	91 h	77 c	61 b	44 b	5 ab	-	-12 a	23 abcde	15 abc
Glyphosate + metsulfuron	2.24 + 0.063	14 ab	65 efg	97 d	100 c	92 c	93 d	-	91 d	52 def	29 bc
Glyphosate + imazapyr	2.24 + 0.84	19 ab	72 fgh	100 d	100 c	100 c	94 d	-	92 d	57 ef	41 c
Metsulfuron + imazapyr	0.063 + 0.84	7 a	42 bcd	100 d	100 c	95 c	91 cd	-	80 d	48 cdef	37 c
Coefficient of variation		2.014	2.014	2.014	2.014	2.014	2.014		2.014	2.014	2.014
Least significant difference (0.05)		16	21	12	21	17	22		25	44	41

[a] Means followed by the same letter are not significantly different at P<0.05. Negative values indicate increase in fern cover above pre-treatment levels

[b] Months after treatment (MAT).

[c] For all treatments except the untreated control, "Induce" surfactant was added at the rate of 0.025% v/v (Helena Chemical).

[d] Year of study initiation.

[e] Plots were mechanically cleared by timber harvest event. Treatment differences remained obscured until 24 MAT.

Table 3-3. Change in Japanese climbing fern foliar cover on untreated control plots from two herbicide trials in a North Florida slash pine plantation

Trial year	Initial % cover [a]	Final % cover [a]	Time period	%Change/year[b]
2002	77.5 (±6.6)	83.4 (±7.4)	3 years	+2.5% NS
2004	70 (±13.7)	72.5 (±10.8)	1 year	+3.6% NS
Mean % increase in climbing fern foliar cover = +3.1% NS				

[a] Mean (n=4), ± standard error

[b] % Change/year = [(Final%-Initial%)/Initial%]/Time period. "NS" indicates non-significant change in cover (SAS Version 9.1, analysis of variance, alpha=0.05)

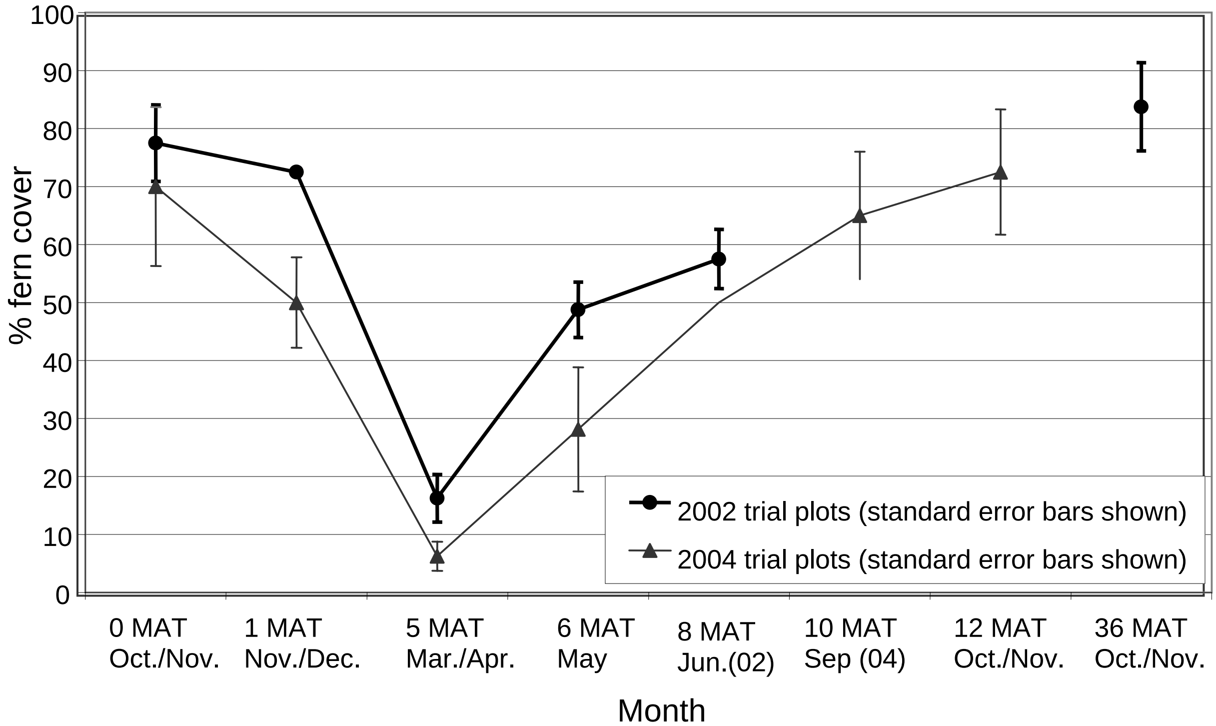

Figure 3-1. Average live foliar cover of Japanese climbing fern on untreated control plots established in October 2002 and November 2004 as part of a multiple herbicide trial in a North Florida pine plantation. Approximate month after treatment (MAT) indicated.

Table 3-4. Japanese climbing fern herbicide treatment options for land managers, based on results of evaluation in a North Florida slash pine plantation, 2002 to 2005.

Recommended herbicides	Solution rates	Treatment effect duration[a] 6 month	10 month	12 month	24 month	Cost per hectare (acre)[b]	Cost per unit[b]
Glyphosate	2.50%	Yes	Yes	Yes	Yes	\$15 to \$43 (\$6 to \$17)	\$12 to \$35 per gallon
Glyphosate	5%	Yes	Yes	Yes	Yes[c]	\$30 to \$86 (\$12 to \$35)	\$12 to \$35 per gallon
Glyphosate + imazapyr	2.5% + 0.94%	Yes	Yes	Yes	Yes[c]	\$153 (\$62)	
Glyphosate + metsulfuron	2.5% + 0.56 g/L (0.075 oz/gal)	Yes	Yes	Yes	Yes[c]	\$111 (\$45)	
Imazapyr	0.94%	Yes	Yes	Yes	Yes	\$116 (\$47)	\$250 per gallon
Metsulfuron+ imazapyr	0.56 g/L (0.075 oz/gal) + 0.94%	Yes	Yes	Yes	Yes	\$190 (\$77)	
Metsulfuron	0.56 g/L (0.075 oz/gal)	Yes	Yes	No	No	\$74 (\$30)	\$20 per ounce
Imazapic	1.25%	Yes	Yes	No	No	\$346 (\$140)	\$560 per gallon
Sulfometuron	1.12 g/L (0.15 oz/gallon)	Yes	No	No	No	\$229 (\$93)	\$500 per pound

[a] "Yes" indicates treatment sustained a minimum of 70% control for 6,10,&12 month periods, and a significant difference from control for 24 month period.
[b] based on per unit costs from Ferrell et al. 2006
[c] Greater than 90% control.

CHAPTER 4
CONCLUSIONS AND RECOMMENDATIONS FOR FUTURE RESEARCH

The continual introduction of new species and the expanding distributions of those already present in the United States make the need for objective priority-based management increasingly important (Richardson 2004). However, for many species, the type and scale of economic and environmental impacts are poorly or loosely quantified, making it difficult to prioritize species for use of limited management dollars. The disparity in recognition and prioritization of management of the two non-native invasive climbing ferns in Florida provides an example of the influence of documented or clear ecological impacts (as undeniably seen with Old World climbing fern), versus less obvious, or un-quantified impacts (as seen with Japanese climbing fern).

Further pursuit of site classification data for Japanese climbing fern is not strongly recommended. In general, the successful design and implementation of broad classification assessments is difficult, and identification of meaningful relationships is often not feasible, or efficiently accomplished. In future consideration of a broad site classification study design, if a similar grid system is used, increasing the sampling intensity (i.e. establishing a greater number of transects and plots per study area) might capture a more normal distribution in plot variable values, and Japanese climbing fern cover, allowing for more robust and productive analyses, and possibly more definitive results. Another approach would be to narrow the plot selection to transition zones or "ecotones" between Japanese climbing fern presence and absence which might explain the basic limiting factors that exclude Japanese climbing fern from some areas. The apparent causes are linked to soil inundation. However, the results of the site assessment described in Chapter 2, and review of current distribution ecology in Chapter 1 indicate that it is likely that Japanese climbing fern will occupy a broad range of sites of mesic to hydric moisture regimes, with other site characteristics playing minor and varying effects on establishment, or percent cover.

Some aspects of management of Japanese climbing fern infestations do require further research review. Most invasive species research seeks to answer at least one of the following key questions: “Where does it occur?”, “What are the ecological impacts?”, “How do we manage it and what are the costs?”, and “Can we halt further spread, and how?” Though the results reported herein do begin to address parts of these questions, further work is needed in several areas. Research recommendations to further answer these questions follow.

Ecological Impacts

Of highest research priority is an assessment of the impacts of Japanese climbing fern invasion at the site, species, and community levels. In the United States, Japanese climbing fern has already invaded a significantly larger geographic area than Old World climbing fern, but without the consistent formation of canopy-smothering infestations. Though not extending over the tops of overstory trees, ground-level Japanese climbing fern infestations can reach 70% to 100% groundcover in heavily infested sites. These heavy infestations can effectively preclude recruitment of native plant species, in strata ranging from groundcover to overstory, and in addition may have direct competitive effects on mature plants from midstory to ground level. The impacts of infestations occurring at lower densities over many acres are not as clear, particularly since the ecological functions and importance of individual native species are poorly understood in many cases. Research is needed to evaluate the true impacts of these infestations on native flora and fauna to help clarify the priority of prevention and management of this species on public and private lands. Specific research priorities include: impacts of Japanese climbing fern invasion on native plant communities at groundcover, understory, midstory, and overstory levels; and impacts on native arthropod communities.

Distribution and Spread

Efforts are currently in place through the cooperative Central Florida Lygodium Strategy to evaluate the effect of the 2004 and 2005 hurricane seasons on both the range extension, and level of site occupancy of both climbing ferns (Serbesoff-King 2006). As Japanese climbing fern continues to expand its distribution in central and south Florida, seasonal climatic restrictions will be removed and the relative biological potential of the two species will be more clearly comparable. However, Japanese climbing fern also continues northward range extension, under-recognized by many.

The true extent of Japanese climbing fern’s county-level range in Georgia has been under-reported by the large reporting databases, as verified by queries of individual herbaria in the Southeast. In particular, the curator of the Valdosta State University Herbarium added seven new Georgia county records (Figure 1-3) in a one-month period after simply increasing attention to Japanese climbing fern occurrence in his daily travels, in response to a query from the author (R. Clark, personal communication, 7/14/2006). A similar trend for under-reporting this species probably exists in Alabama, Mississippi, Arkansas, South Carolina, and East Texas. Well beyond simple incursions into the southern regions of infested states, Japanese climbing fern already extends throughout Louisiana, and reaches well into central regions of Mississippi, Alabama, Georgia, and South Carolina.

Recommendations for future work in this area include: active pursuit of an increase in recognition and reporting of Japanese climbing fern occurrence in these states to further aid in understanding the current and future impacts of this species on mesic and hydric natural communities throughout the Southeastern United States. This may require regional or state-level funding for surveys in target areas. In addition to location, descriptions of site should be reviewed for commonalities indicative of common vectors or dispersal mechanisms. Assessment of regional spore loading levels in Northwest Florida and in the central and northern ranges of each infested state in the Southeastern United States (Figure 1-3) may help clarify the infestation extent and eradication feasibility across the current range. Spore loading assessments must be able to differentiate spores of *L. japonicum* and *Lygodium palmatum.*

Outside the United States, the present status and possible future distribution of Japanese climbing fern and Old World climbing fern in the Caribbean islands may also need review. These systems may be particularly vulnerable due to the fact that an "empty niche" may exist in Caribbean forests due to the dearth of native vines in those systems (Horvitz et al. 1998), and proximity to the heavily infested State of Florida (Figure 1-2).

Management Gaps

Rejmanek and Richardson (1996) suggest that non-native species become successful invaders based on small widely dispersed seeds becoming established in anthropogenically-disturbed ecosystems. Horovitz et al. (1998) propose that invasion success is determined by recruitment and persistence at low densities in natural habitats, followed by rapid population expansion after a disturbance event. Both of these invasion paradigms apply to Japanese climbing fern, indicating that the scattered, lower density populations typical of many infested sites are simply the preliminary stages of infestations which await release through site or canopy disturbance. In forested sites under silvicutural management, the unfortunate result may be rapid conversion of scattered, incipient populations to site dominating near-monocultures following full or partial harvests. This conversion will complicate future management, including increasing the cost and reducing the success of reforestation efforts, and altering the form and diversity of groundcover plant communities. This effect has already been observed following removal of pine needle litter from mesic pine plantations in north Florida as a component of the pine straw industry (Van Loan 2001).

In particular because of Japanese climbing fern's reproductive strategy, proactive treatment at the invasion "front" is a far better management strategy than waiting until regional populations and spore loading prevent eradication from being a feasible management goal. Therefore, development of reporting programs and supporting training sessions on identification and treatment of Japanese climbing fern in the counties forming the northern extent of the species in the United States is recommended. State and county government involvement is critical to raise public awareness, and regional experts will be required to verify sites as necessary. Public and private land managers should receive focused training and land management funding support, concurrent with provision of the same to counties immediately adjacent to the north and south. This way, further northward spread of the species may be halted, allowing focused local education and treatment programs to begin moving the range southward. This strategy is recommended for immediate implementation in the Southeastern United States. Eradication may be possible in some regions or states, leaving heavily infested states with pockets requiring ongoing maintenance-control treatments.

Given that site-based restrictions on establishment are potentially limited beyond general site hydrology, improving management of infested sites may be far more important targets for future research than further assessment of habitat preferences. Some recommended priorities include: assessment of the effect of repeated herbicide applications of higher-performing herbicide treatments within a single year, and over several years. A comparison of treatment efficacy and associated seasonal spore loading in stands treated in late spring versus fall may further assist in setting treatment timelines. Given the fact that one single treatment will not eradicate this species from a site, quantification of the extent and cost of an eradication-based management strategy will aid land managers in long term planning. The effect of varying levels and timings of site disturbance should be assessed, including common silvicultural treatments to facilitate adaptive management on managed lands. When establishing herbicide or other treatment plots in Japanese climbing fern infestations, a minimum plot size of 3.4 m by 6.8 m to prevent treatment effects from being carried into adjacent plots. A second approach would be to use buffers between plots

This leads to the next recommendation; focused pursuit of a biological control agent for Japanese climbing fern. Presently, Japanese climbing fern receives some review as a result of the search for an agent for Old World climbing fern. Range overlap with *L. palmatum* does reduce the likelihood that a host-specific agent will be found, but the possibility does still exist. Foreign exploration for possible insect or fungal agents in various areas of the range of Japanese climbing fern is recommended to address the feasibility of biological control of Japanese climbing fern.

Regulatory Gaps

Finally, increased recognition of this plant at a regulatory level may be the only path to halting its continued spread. In particular, an in-state review of the noxious weed status of this species in Georgia, Texas, Mississippi, Arkansas, Lousiana, South Carolina, and North Carolina is highly recommended, and may aid in generating adequate focus to effectively treat and manage the widely occurring incipient populations that reflect Florida infestation levels of decades past. Concurrently, the possible range in the United States should be reviewed more carefully, as this is a key question to support petition of this species for federal noxious weed status. If at minimum, the Southeastern Coastal Plain represents the possible United States range, populations are not yet known in: Tennessee, Missouri, Oklahoma, or Virginia, as well as large areas of eastern Texas, and northern Mississippi (Figure 1-2). If however, the possible range is closer to that indicated by the current northern (43º N) and southern (33º S) extent of the species, appropriate sites ranging from Boston, Massachusetts, USA to Santiago, Chile might be vulnerable (Figure 1-2). Both scenarios are possible at some level, and both support the need for designation of Japanese climbing fern as a federal noxious weed by the United States Department of Agriculture (USDA). Development of a petition to this effect for submission to the USDA is recommended.

The need for coordination of regulatory and management approaches is easily illustrated by forest product movement and the related management practices; and the potential for spread of Japanese climbing fern associated with each. The ability for Japanese climbing fern to be baled and sold in contaminated pinestraw bales has been shown repeatedly, indicating that despite some legal basis an effective preventative, or punative system has not yet been implemented. The most comprehensive approach to preventing the continued vectoring of Japanese climbing fern this way is to halt the continued harvest or sale of any forest products from infested stands. Legal basis exists already for this action in Florida and Alabama. However, comprehensive implementation of such a requirement would require significant increases in funding for state or federal agricultural inspectors in rural, forested areas. One approach which may achieve practical success would include funding and training a rapid response team in each state with known infestations to address further expansion, particularly northward. A second step would included providing cost-share funds for treatment of infestations on private forestlands. With participation in the cost-share program, landowners could provide forest products through a “Fern-free” certification program, and successfully comply with state agricultural rules.

As the gaps in knowledge continue to be filled with current and future research, a more complete understanding of Japanese climbing fern’s occurrence and impacts in the Southeastern United States will be possible. Communication of the real nature of these impacts will be key in successful future management of this species, and the natural communities vulnerable to its invasion. The results reported in this document represent early steps in development of a regional landscape-based management approach for this species capable of effectively addressing its regional spread.

APPENDIX
INITIAL ANALYSIS TABLES FROM CHAPTER 2

Table A-1. Abiotic plot variable with significant relationship to presence or absence of Japanese climbing fern (*Lygodium japonicum*) in three North Florida forests.

Variable[a]	*Lygodium* status	Plots	Mean	Std. error	Kruskal-Wallis values[b]	Spearman R correlation coefficients[c]	Correlation strength[d]
Site	Present	29	2.52	0.12	19.42 (P<0.0001)*	0.50 (P<0.0001)*	Moderate
	Absent	46	1.67	0.11			
Plot PAR	Present	29	133.49	34.75	15.00 (P<0.0001)*	-0.45 (P<0.0001)*	Weak
	Absent	46	454.98	53.59			
Hydrology class	Present	29	2.59	0.12	8.57 (P=0.003)*	0.34 (P=0.003)*	Weak
	Absent	46	1.96	0.14			
Plot flood duration	Present	29	2.07	0.42	4.74 (P=0.029)*	0.25 (P=0.029)*	Weak
(months, 2003)	Absent	46	1.03	0.26			
Percent flooded	Present	29	0.27	0.07	4.51 (P=0.034)*	0.25 (P=0.033)*	Weak
	Absent	46	0.15	0.05			

[a] PAR=photosynthetically active radiation.
[b] Asterisk* indicates significant value – Kruskal-Wallis one way analysis of variance test p-value, alpha = 0.05, SAS Version 9.1
[c] Asterisk* indicates significant value - Spearman's correlation coefficient and p-values, alpha=0.05, SAS Version 9.1
[d] Correlation strength: weak=0.2-0.49, moderate=0.5-0.79, strong=0.8-1.0

Table A-2. Biotic plot variables with significant non-paramteric relationships to presence or absence of Japanese climbing fern (*Lygodium japonicum)* in three North Florida forests.

Variable [a]	*Lygodium* status	Plots	Mean	Standard error	Kruskal-Wallis values[b]	Spearman R correlation coefficients[c]	Correlation strength[d]
Plot % understory cover	Present	29	29.14	4.49	11.30 (P<0.0001)*	-0.39 (P=0.0005)*	Weak
	Absent	46	61.30	5.85			
Plot % canopy cover	Present	29	60.04	2.56	10.48 (P<0.001)*	0.38 (P=0.0009)*	Weak
	Absent	46	41.34	3.38			
Plot tree evenness index	Present	29	0.65	0.07	8.71 (P=0.003)*	0.32 (P=0.005)*	Weak
	Absent	44	0.37	0.06			
Plot tree species richness	Present	29	3.07	0.30	6.72 (P=0.010)*	0.30 (P=0.009)*	Weak
	Absent	46	2.13	0.23			
Plot tree diversity index	Present	29	2.24	0.22	6.97 (P=0.008)*	0.31 (P=0.074)*	Weak
	Absent	44	1.72	0.15			
Trees per plot	Present	29	11.07	0.98	4.25 (P=0.040)*	0.24 (P=0.038)*	Weak
	Absent	46	8.37	0.94			

[a] Plot tree evenness index = Pielou J', plot tree diversity index=Simpson's reciprocal 1/D.

[b] Asterisk* indicates significant value – Kruskal-Wallis one way analysis of variance test p-value, alpha = 0.05, SAS Version 9.1

[c] Asterisk* indicates significant value – Spearman R correlation coefficient, alpha=0.05, SAS Version 9.1

[d] Correlation strength: weak=0.2-0.5, moderate=0.5-0.8, strong=0.8-1.0

Table A-3. Soil variables with significant parametric relationship to presence or absence of Japanese climbing fern (*Lygodium japonicum)* in three North Florida forests.

Variable (data transformation)[a]	*Lygodium* status	N	Mean	Standard error	T values[b]	Pearson correlation coefficients[c]	Correlation strength[d]
Soil pH (inverse)	Present	29	5.99	0.12	5.17 (P<0.0001)*	0.51 (P<0.0001)*	Moderate
	Absent	46	5.23	0.09			
Soil P (inverse)	Present	29	26.03	2.78	5.24 (P<0.0001)*	0.51 (P<0.0001) *	Moderate
	Absent	46	9.41	1.96			
Soil Ca (natural log)	Present	29	283.93	35.51	-5.72 (P<0.0001)*	0.49 (P<0.0001)*	Weak
	Absent	46	101.26	20.32			
Soil Fe (inverse)	Present	29	13.52	1.14	-5.18 (P<0.0001)*	-0.37 (P=0.001)*	Weak
	Absent	46	31.50	4.08			
Soil Mg (inverse)	Present	29	34.03	3.61	5.04 (P<0.0001)*	0.38 (P=0.0007)*	Weak
	Absent	46	17.96	2.81			
Soil Al	Present	29	191.71	18.38	4.17 (P<0.0001)*	-0.39 (P=0.0005)*	Weak
	Absent	46	332.89	28.41			
Soil K (natural log)	Present	29	40.76	7.02	-2.90 (P=0.005)*	0.32 (P=0.005)*	Weak
	Absent	46	21.27	2.89			
Soil Zn	Present	29	2.19	0.30	-2.48 (P=0.017)*	0.30 (P=0.009)*	Weak
	Absent	46	1.33	0.16			
Soil Na	Present	29	29.56	6.73	-2.28 (P=0.028)*	0.29 (P=0.011)*	Weak
	Absent	46	13.03	2.68			

[a] Where indicated, data transformations utilized to improve distribution normality for parametric analyses.
[b] Asterisk* indicates significant value - two sample T-test, alpha = 0.05, SAS Version 9.1
[c] Asterisk* indicates significant value - Pearson's correlation coefficient, alpha=0.05, SAS Version 9.1
[d] Correlation strength: weak=0.2-0.5, moderate=0.5-0.8, strong=0.8-1.0

Table A-4. Tree species with importance values (IV) significantly related to presence or absence of Japanese climbing fern (*Lygodium japonicum*) in three North Florida forests.

Species	Veg. index[a]	Total plots	*Lygodium* status	Plots	Mean IV	Kruskal-Wallis values[b]	Spearman R correlation coefficients[c]	Corr. Strength[d]
Liquidambar styraciflua L.	FAC	10	Present	10	25.4	17.94 (P=0.0001)*	0.34 (P=0.003)*	Weak
			Absent	0	5.0			
Pinus palustris Mill.	FACU	28	Present	3	28.3	-15.88 (P<0.0001)*	-0.45 (P<0.0001)*	Weak
			Absent	25	108.1			
Carya aquatica (Michx. f.) Nutt.	OBL	16	Present	12	40.2	11.88 (P=0.0006)*	0.37 (P=0.001)*	Weak
			Absent	4	13.9			
Quercus lyrata Walt.	OBL	9	Present	7	13.1	6.28 (P=0.012)*	0.19 (P=0.10)	
			Absent	2	6.6			
Ulmus americana L.	FACW	9	Present	7	13.3	6.28 (P=0.012)*	0.19 (P=0.11)	
			Absent	2	7.4			
Carpinus caroliniana Walt.	FACW	3	Present	3	5.6	4.89 (P=0.027)*	0.22 (P=0.06)	Weak
			Absent	0	2.0			
Quercus nigra L.	FACW	14	Present	9	17.8	4.35 (P=0.037)*	0.29 (P=0.01)*	Weak
			Absent	5	9.7			
Taxodium distichum (L.) L. C. Rich	OBL	5	Present	4	10.1	4.00 (P=0.046)*	0.27 (P=0.02)*	Weak
			Absent	1	3.3			

[a] Vegetative index: FAC = facultative, FACU = facultative upland, OBL=obligate wetland, FACW=facultative wetland.
[b] Asterisk* indicates significant value - Kruskal-Wallis analysis of variance p-values, alpha = 0.05, SAS Version 9.1
[c] Asterisk* indicates significant value - Spearman R correlation coefficient, alpha=0.05, SAS Version 9.1
[d] Correlation strength: weak=0.2-0.5, moderate=0.5-0.8, strong=0.8-1.0

Table A-5. Tree species with importance values (IV) not significantly related to presence or absence of Japanese climbing fern (*Lygodium japonicum*) in three North Florida forests.

Species	Vegetative index[a]	Total plots	*Lygodium* status	Plots	Mean IV	Std. dev. IV
Pinus elliottii Engelm.	FACW	18	Present	8	58.2	83.2
			Absent	10	41.0	67.7
Nyssa aquatica L.	OBL	8	Present	5	20.0	40.6
			Absent	3	15.3	44.1
Nyssa sylvatica Marsh.	OBL	6	Present	1	4.8	9.7
			Absent	5	13.5	35.3
Acer rubrum L.	FACW	6	Present	3	7.0	16.2
			Absent	3	9.6	29.4
Betula nigra L.	OBL	5	Present	4	8.8	15.6
			Absent	1	4.3	8.9
Quercus stellata Wangenh.	FACU	5	Present	0	3.0	0
			Absent	5	8.7	20.3
Quercus falcata Michx.	FACU	4	Present	0	2.0	0
			Absent	4	5.5	14.3
Magnolia grandiflora L.	FAC	3	Present	0	2.0	0
			Absent	3	4.8	13.2
Liriodendron tulipifera L.	FACW	3	Present	1	5.2	17.5
			Absent	2	4.4	12.0
Quercus laevis Walt.	FACW	3	Present	1	2.6	3.3
			Absent	2	2.6	2.7
Bumelia lycioides (L.) Pers.	FAC	2	Present	0	1.0	0
			Absent	2	1.4	1.9
Persea palustris (Raf.) Sarg.	OBL	2	Present	0	1.0	0
			Absent	2	1.5	2.4
Quercus hemisphaerica Bartr. ex Willd.	FAC	2	Present	0	1.0	0
			Absent	2	2.6	7.8
Taxodium ascendens Brongn.	OBL	2	Present	0	1.0	0
			Absent	2	1.5	2.4
Catalpa bignonioides Walt.	FAC	1	Present	1	1.3	1.9
			Absent	0	1.0	0
Celtis laevigata Willd.	FACW	1	Present	1	2.9	10.4
			Absent	0	1.0	0
Ligustrum sinense Lour.	FAC	1	Present	1	4.1	16.9
			Absent	0	1.0	0
Nyssa ogeche Bartr ex Marsh.	OBL	1	Present	1	3.6	14.1
			Absent	0	1.0	0
Planera aquatica Walt. ex Gmel.	OBL	1	Present	1	2.2	6.3
			Absent	0	1.0	0
Platanus occidentalis L.	FACW	1	Present	1	2.2	6.5
			Absent	0	1.0	0

Table A-5. Continued

Species	Vegetative index[a]	Total plots	*Lygodium* status	Plots	Mean IV	Std. dev. IV
Populus heterophylla L.	OBL	1	Present	1	1.3	1.9
			Absent	0	1.0	0
Quercus michauxii Nutt.	FACW	1	Present	1	2.5	8.0
			Absent	0	1.0	0
Viburnum nudum L.	FACW	1	Present	1	1.8	4.5
			Absent	0	1.0	0
Cornus florida L.	FACU	1	Present	0	1.0	0
			Absent	1	1.3	2.1
Cyrilla racemiflora L.	FAC	1	Present	0	1.0	0
			Absent	1	1.5	3.7
Ilex opaca Ait.	FAC	1	Present	0	1.0	0
			Absent	1	1.0	0.3
Diospyros virginiana L.	FAC	1	Present	0	1.0	0
			Absent	1	1.6	4.3
Lindera benzoin	FACW	1	Present	0	1.0	0
(L.) Blume			Absent	1	1.5	3.1
Myrica cerifera L	FAC	1	Present	0	1.0	0.2
			Absent	1	1.2	1.0
Persea borbonia	FAC	1	Present	0	1.0	0
(L.) Spreng.			Absent	1	1.3	1.8
Pinus taeda L.	FAC	1	Present	0	1.0	0
			Absent	1	3.3	15.5
Quercus incana Bartr.	FACU	1	Present	0	1.0	0
			Absent	1	1.3	2.2
Symplocos tinctoria	FAC	1	Present	0	1.0	0
(L.) L' Her.			Absent	1	2.4	9.7
Gleditsia aquatica	OBL	1	Present	1	1.7	3.5
Marsh.			Absent	1	1.6	4.1
Magnolia virginiana L.	OBL	1	Present	0	3.7	14.3
			Absent	1	1.3	1.6

[a] Vegetative index: FAC = facultative species, FACU = facultative upland species, FACW=facultative wetland species, OBL=obligate wetland species.

LIST OF REFERENCES

Aherns, W. H. ed. 1994. Herbicide Handbook seventh edition. Champaign, IL: Weed Science Society of America. 352 p.

Allen, H. L., T. R. Fox, and R. G. Campbell. 2005. What is ahead for intensive pine plantation silviculture in the south? South. J. Appl. For. 29:62–69.

Alston, A. H., and R. E. Holtum. 1959. Notes on the taxonomy and nomenclature in the genus *Lygodium* (Schiziaceae). Reinwardtia 5:11–22.

Barnard, E. L. and A. N. Van Loan. 2002. An assessment of fusiform rust and selected non-native invasive pest plants in longleaf and slash pine plantations established with Florida Division of Forestry seedlings. A report to the USDA Forest Service Forest Health Monitoring Program, Region 8, Atlanta, GA: Florida Division of Forestry. 17 p.

Beckner, J. 1968. *Lygodium microphyllum*, another fern escaped in Florida. Am. Fern J. 58:93–94.

Berquist, G. T., A. M. Pable, M. C. Killingsworth, and J. A. Silvestri. 1995. Apalachicola River and Bay ecosystem management plan: environmental indicator system. Florida Center for Public Management, Tallahassee, FL: Florida State University. 19 p.

Betrock Information Systems. 2002. Betrock's PlantFinder: wholesale guide to foliage and ornamental plants. Hollywood, FL: Betrock's Information Systems, Inc. 148 p.

Brandt, L.A., and D.W. Black. 2001. Impacts of the introduced fern, *Lygodium microphyllum* on the native vegetation of tree islands in the Arthur R. Marshall Loxahatchee National Wildlife Refuge. Florida Scientist 64:191–196.

Brown, M. J. 1995. Florida's Forests, 1995. Resour. Bull. SRS-48. Asheville, NC: U. S. Department of Agriculture, Forest Service, Southern Research Station. 83 p.

Brown, R. B., E. L. Stone, and V. W. Carlisle. 1990. Soils., *In* R. L. Myers, and J. J. Ewel eds. 1990 Ecosystems of Florida. Orlando, FL: University of Central Florida Press. Pp. 35–100.

Brumback, W. E. 1985. Raising the climbing fern from spores. Arnoldia 45:27–29.

Burke, M. J., and J. P. Grime. 1996. An experimental study of plant community invisibility. Ecology 77:776–1009.

Chornesky, E. A., A. M. Bartuska, G. H. Aplet, K. O. Britton, J. Cummings-Carlson, F. W. Davis, J. Eskow, D. R. Gordon, K. W. Gottschalk, R. A. Haack, A. J. Hansen, R. N. Mack, F. J. Rahel, M. A. Shannon, L. A. Wainger, and T. B. Wigley. 2005. Science priorities for reducing the threat of invasive species to sustainable forestry. BioScience 55:335–348.

Clawson, R. J., B. G. Lockaby, and B. Rummer. 2001. Changes in production and nutrient cycling across a wetness gradient within a floodplain forest. Ecosystems 4:126–138.

Clewell, A. F. 1985 Guide to the Vascular Plants of the Florida Panhandle. Tallahassee, FL: University Press of Florida. 605 p.

Cohen, J. 1988. Statistical power analysis for the behavioral sciences (2nd ed.). Mahwah, NJ: Lawrence Erlbaum Assoc. 500 p.

D'Antonio, C. D. and L. A. Meyerson. 2002. Exotic plant species as problems and solutions in ecological restoration: a synthesis. Restoration Ecol. 10:703–713.

Darke, A. K., and M. R. Walbridge. 2000. Al and Fe biogeochemistry in a floodplain forest: implications for P retention. Biogeochemistry 51:1–32.

Davis, M. A., J. P. Grime, and K. Thompson. 2000. Fluctuating resources in plant communities: a general theory of invisibility. J. Ecol. 88:528–534.

Diggle, A. J., P. B. Neve, and F. P. Smith. 2003. Herbicides used in combination can reduce the probability of herbicide resistance in finite weed populations. Weed Res. 43:371–382.

Dixon, J. B., and D. G. Schulze. 2002. Soil mineralogy with environmental applications. Madison, WI: Soil Science Society of America Pp.1–15.

Duryea, M. 1998. Pine straw management in Florida's forests, Circular 831. Gainesville, FL: Florida Cooperative Extension Service, IFAS, University of Florida. 12 p.

Dyer, A. F., and S. Lindsay. 1992. Soil spore banks of temperate ferns. Am. Fern J. 82:89–122.

Evans, C. and D. Moorhead. 2005. Georgia EPPC events highlight emerging threats. Wildland Weeds 8:4–5.

Ezell, A. W., and L. Nelson. 2001. Weed control and crop tolerance after pre-emergent and post-emergent applications of sulfometuron in oak (*Quercus* spp.) plantations. Weed Tech. 15:585–589.

[FLDEP] Florida Department of Environmental Protection. 2006. Florida Department of Environmental Protection's Wetland Delineation Vegetative Index : Web page: www.dep.state.fl.us/water/wetlands/delineation/vegindex/vegindex.htm. Accessed: July 10, 2006.

[FLEPPC] Florida Exotic Pest Plant Council. 2005. Exotic Pest Plant Database: Web page: www.fleppc.org/database/data_intro.htm. Accessed: December, 1, 2005.

[FLEPPC] Florida Exotic Pest Plant Council. 2005. Florida Exotic Pest Plant Council's 2005 List of Invasive Species: Web page: www.fleppc.org. Accessed: December, 10, 2005.

[FNAI] Florida Natural Areas Inventory. 2005. Florida Invasive Plants GeoDatabase - June 2005, Web page: www.fnai.org/gis_data.cfm. Accessed: December 10, 2005.

Fan, W. 1996. A manual of Chinese herbal medicine: principles and practice of easy reference. Boston, MA: Shambhala Publishing. p. 207.

Ferrell, J. A., C. J. Gray, and G. E. MacDonald. 2006. Approximate Herbicide Pricing 2006, SS-AGR-16. Gainesville, FL: Florida Cooperative Extension Service, University of Florida. 4 p.

Ferriter, A. ed. 2001. *Lygodium* Management Plan for Florida, a report from the Florida Exotic Pest Plant Council's *Lygodium* Task Force. West Palm Beach, FL: South Florida Water Management District. 59 p.

Ferriter, A. and Pernas, T. 2006. An explosion in slow motion: tracking the spread of *Lygodium microphyllum* in Florida. Wildland Weeds 9:7–9.

Foxcroft, L. C., D. M. Richardson, M. Rouget, and S. MacFayden. 2004. Reconstructing fifty years of Opuntia stricta invasion in the Kruger National Park: environmental determinants and propagule pressure. Divers. Distrib. 10:427–437.

Gias Uddin, M.D, M. Mahal, and M.K. Pasha. 1997. Taxonomic account of genus *Lygodium* Sw. (Lygodiaceae) in Bangladesh. Indian Fern Journal 14:33–37.

Gordon, D.R. 1998. Effects of invasive, non-indigenous plant species on ecosystem processes: lessons from Florida. Ecol. Appl. 8:975–989.

Hamilton, R. G.. 1988. The significance of spore ibanks in natural populations of *Athyrium pycnocarpon* and *A. thelypteroides.* Am. Fern J. 78:96–104.

Haywood, J.D., F.L. Harris, H.E. Grelen, and H.A. Pearson. 2001. Vegetative response to 37 years of seasonal burning on a Louisiana longleaf pine site. S. J. App. For. 25:122–130.

Hipps, C. B. 1989. Japanese climbing fern: an exotic vine for a shady fence or trellis. Horticulture, (August 1989):38–39.

Hodges, A., D. Mulkey, J. Alavalapati, D. Carter, and C. Kiker. 2004. Executive summary: Economic impacts of the forest products industry in Florida. Florida Forests 3:12–15.

Holttum, R. E. 1959. Flora Malesiana, Series II, Pteridophyta. Volume I. Leyden, The Netherlands: Noordhoff International Publishing. Pp. 44–51.

Horvitz, C. C., and A. Koop. 2001. Removal of non-native vines and post-hurricane recruitment of tropical hardwood forests of Florida. Biotropica 33:268-281.

Horovitz, C.C., J.B. Pascarella, S. McMann, A. Freedman, and R.H. Hofstettler. 1998. Functional roles of invasive, non-indigenous plants in hurricane-affected subtropical hardwood forests. Ecol. Appl. 8:947–974.

Hoshizaki, B. J. and R. C. Moran. 2001. Fern Growers Manual. Portland, OR: Timber Press 624 p.

Hutchinson, J. T., A. Ferriter, K. Serbesoff-King, K. Langeland, and L. Rodgers. 2006. Old World climbing fen management plan for Florida: A report from the Florida Exotic Pest Plant Council's Lygodium Task Force. Second Edition. West Palm Beach, FL: Florida Exotic Pest Plant Council Lygodium Task Force. 109 p.

Hutchinson, J. T. and K. A. Langeland. 2006. Potential spread of *Lygodium microphyllum* spores by herbicide applicators. Wildland Weeds 9:13–15.

Jones, K. A. M. B. Rayamajhi, P. D. Pratt, and T. K. Van. 2003. First report of the pathogenicity of *Colletotrichum gloeosporioides* on invasive ferns, *Lygodium microphyllum*, and *L. japonicum*, in Florida. Plant Dis. 87:101.

Kagawa, T. and M. Sugai. 1991. Involvement of gibberellic acid in phytochrome-mediated spore germination of the fern Lygodium japonicum. J. Plant Physiol. 138:299–303.

Ko, W. 2003. Germination of fern spores in natural soils. Am. Fern. J. 93:70–75.

Kudsk, P. and J. C. Streibig. 2003. Herbicides: a two-edged sword. Weed Research 43:90–102.

Langeland, K. A., and K. C. Burks. eds. 1998. Identification and Biology of Non-native Plants in Florida's Natural Areas. Gainesville, FL: University of Florida Press. Pp. 14–15.

Lauer, D. K., H. E. Quicke, and P. J. Minogue. 2002. Early season site preparation with different imazapyr formulations. Proc. South. Weed Sci. 55:83–88.

Lavorel, S., A. H. Prieur-Richard, and K. Grigulis. 1999. Invasibility and diversity of plant communities: from patterns to processes. Divers. Distrib. 5:41–49.

Lee, D. J.. 2005. The private cost of upland invasive plants in Florida, University of Florida, Institute of Food and Agricultural Sciences, Florida's Invasive Species Working Group: Web page: http://www.iswgfla.org/. Accessed: July, 15, 2006.

Leitman, H. M., J. E. Sohn, and M. A. Franklin. 1983. Wetland hydrology and tree distributions of the Apalachicola River floodplain, Florida, Water Supply Paper 2196. Tallahassee, FL: U. S. Geological Survey. 52 p.

Lloyd, R.M., and E.J. Klekowski. 1970. Spore germination and viability in pteridophyta: evolutionary significance of chlorophyllous spores. Biotropica 2:129–137.

Lockhart, C. 2006. The double-trouble ferns: status surveys of *Lygodium* treatment sites. Wildland Weeds 9:23–24.

Londo, A. J., J. D. Kusha, and R. C. Carter. 2006. Soil pH and tree species suitability in the South, SREF-FM-002. Athens, GA: Southern Regional Extension Service. 4 p.

Lott, M. S., J. C. Volin, R. W. Pemberton, and D. F. Austin. 2003. The reproductive biology of the invasive ferns *Lygodium microphyllum* and *L. japonicum* (Schizaeaceae): implications for invasive potential. Am. J. Bot. 90:1144–1152.

Louisiana State University. 2006. The Herbarium of Louisiana State University, accession database: Web page: http://www.herbarium.lsu.edu. Accessed: May 27, 2006.

Magurran, A. E.. 1988. Ecological diversity and measurement. Princeton, NJ: Princeton University Press. 179 p.

Marthani, G. P., V.K. Bahuguna, and P. Lal. Effect of forest fires on the ground vegetation of the moist deciduous sal forest. India Forester 112:646–667.

Martin, W. H., S. G. Boyce, and A. C. Echternacht eds. 1993. Biodiversity of the southeastern United States: lowland and terrestrial communities. New York, NY: John Wiley and Sons. 502 p.

Mehrotra, K., C. K. Mohan, and S. Ranka. 2000. Elements of artificial neural networks. Boston, MA: MIT Press. 348 p.

Miller, J.H. 2000. Exotic Pest Plants of Southeastern Forests: a field guide for forest inventory and monitoring. Auburn: USDA Forest Service, Southern Research Station. p. 41.

Miller, J. H. 2003. Nonnative Invasive Plants of Southern Forests: A field guide for forest inventory and monitoring, Gen. Tech. Rep. SRS-62. Asheville, NC: USDA Forest Service, Southern Research Station. 93 p.

Miller, J. H., E. B. Chambliss and C. T. Bargeron. 2004. Invasive plants of the thirteen southern states: Web page: http://www.invasive.org/seweeds.cfm. Accessed: May 22, 2006.

Mueller, R. J. 1981. Developmental morphology of the shoot of *Lygodium (*Schizaeaceae), Ph. D. dissertation. Berkeley, CA: University of California, Berkeley 141 p.

Mueller, R. J. 1982. Shoot morphology of the climbing fern *Lygodium* (Schizaeaceae): general organography, leaf initiation, and branching. Bot. Gaz. 143:319–330.

Mueller, R. J. 1983. Indeterminate growth of the climbing leaves of a fern. BioScience 33:586–587.

Nauman, C. E. 1993. *Lygodium* C Presl. In, Flora of North America Editorial Committee, eds., Flora of North America: Volume 2, Pteridophytes and Gymnosperms. New York:, NY: Oxford University Press. Pp.114–116.

Page, C. N. 2002. Ecological strategies in fern evolution: a neopteridological overview. Rev. Paleobot. Palynol. 119:1–33.

Peck, J., C. Peck and D. Farrar. 1990. Influences of life history attributes on formation of local and distant fern populations. Am. Fern J. 80:126–142.

Pemberton, R. W. and A. P. Ferriter. 1998. Old World climbing fern (*Lygodium microphyllum*), a dangerous invasive weed in Florida. Am. Fern J. 88:165–175.

Pemberton, R. W. 2006. Taking down climbing fern: biological control of *Lygodium microphyllum*. Wildland Weeds 9:10–11.

Pemberton, R. W. 2003. The common staghorn fern, *Platycerium bifurcatum*, naturalizes in southern Florida. Am. Fern. J. 93:203–206.

Penrod, K. A., and L. H. McCormick. 1996. Abundance of viable hay-scented fern spores germinated from hardwood forest soils at various distances from a source. Am. Fern J. 86:69–79.

Perez-Garcia, B. A. Orozco-Segovia, and R. Riba. 1994. The effects of white flourescent light, far-red light, darkness, and moisture on spore germination of *Lygodium heterodoxum* (Schizaeaceae). Am. J. Bot. 81:1367–1369.

Pezeshki, S. R. 2001. Wetland plant responses to soil flooding. Environ. Exp. Bot. 46:299–312.

Pielou, E. C. 1975. Ecological diversity. New York, NY: Wiley and sons. Pp. 7–8.

Pimental, D., L. Lach, R. Zuniga, and D. Morrison. 2000. Environmental and economic costs associated with non-indigenous species in the United States. BioScience 50:53–65.

Platt, W. J., and M. W. Schwartz. 1990. Temperate hardwood forests. *In* R. L. Myers and J. J. Ewel eds. Ecosystems of Florida. Orlando, FL: University of Central Florida Press. Pp. 194–229.

Prantl, K. 1881. Die Schizaeaaceen, Morphology der gefasskryptogamen volume 2. Leipzig: Verlag Von Wilhelm Engelmann. Pp. 7–85.

Prieur-Richard, A. H., and S. Lavorel. 2000. Invasions: the perspective of diverse plant communities. Austral Ecol. 25:1–7.

Punetha, N. 1987. Apical dominance and apical inhibition in the fronds of *Lygodium japonicum* (Thunb.)Sw.. Phytomorphology 37:299–301.

Punetha, N. 2000. Phenology of the climbing leaf of *Lygodium japonicum* (Thunb.) Sw. (Schizaeaceae). Phytomorphology 50:75–85.

Radford, A. E., H. E. Ahles, and C. R. Bell. 1968. Manual of the Vascular Plants of the Carolinas. Chapel Hill, NC: University of North Carolina Press. 1183 p.

Rainbolt, C. R., B. A. Sellers, J. A. Ferrell, and G. E. MacDonald 2005. Herbicide Resistant Weeds, SS-AGR-243. Gainesville, FL: Florida Cooperative Extension Service, University of Florida. 8 p.

Ranal, M. A. 2003. Soil spore bank of ferns in a gallery forest of the Ecological Station of Pangg, Uberlandia, MG, Brazil. Am. Fern J. 93:97–115.

Ranal, M. A.. 2004. Bark spore bank of ferns in a gallery forest of the Ecological Station of Panga, Uberlandia-MG, Brazil. Am. Fern J. 69:57–69.

Rayachhetry, M. B., R. W. Pemberton, L. L. Smith, and R. Leahy. 2001. Pathogenicity assessment of *Puccinnia lygodii*, a potential biological control agent of *Lygodium japonicum* in southeastern United States. Plant Dis. 85:232.

Rhodenbaugh, E. J. and J. L. Yeiser. 1994. Hardwood seedling tolerance to selected Oust treatments. Proc. South Weed Sci. Soc. 47:98–103.

Roberts, D. 1996. Climbing fern wreaks wetland havoc. Florida Department of Environmental Protection Resource Management Notes 8:13.

Roberts, D. 1997. *L. microphyllum* research and mitigation at Jonathan Dickinson State Park. Florida Department of Environmental Protection Resource Management Notes 9:30–32.

Roberts, D. and D. Richardson. 1994. Exotic climbing fern. Florida Department of Environmental Protection Resource Management Notes 6:16.

Royal Botanic Gardens Melbourne. 2006. Australia's Virtual Herbarium: Web page: http://www.rbg.vic.gov.au/cgi-bin/avhpublic/avh.cgi. Accessed: May 27, 2006..

Russell, A. E., J. W. Raich, and P. M. Vitousek. 1998. The ecology of the climbing fern *Dicranopteris linearis* on windward Mauna Loa, Hawaii. J. Ecol. 86:765–779.

Sahi, A. N. and S. K. Singh. 1994. Effect of sulfite of spore germination and rhizoid development in the tropical fern Lygodium japonicum (Filicales: Lygodiaceae). Rev. Biol. Trop. 42:53–57.

[SAS] Statistical Analysis Systems 2005. SAS Version 9.1 Cary, NC: SAS Institute.

Schuler, J. L., D. J. Robison, and H. E. Quicke. 2004. Assessing the use of Chopper herbicide for establishing hardwood plantations in a cutover site. South. J. App. For. 28:163–170.

Shea, K., and P. Chesson. 2002. Community ecology theory as a framework for biological invasions. Trends Ecol. Evol. 17:170–176.

Siegel, S. 1988. Non-parametric Statistics for the Behavioral Sciences (2nd Edition). New York, NY: Castellan Jr. Pp.174–83.

Singh, S. and G. Panigrahi. 1984. Systematics of the genus *Lygodium* Sw. (Lygodiaceae) in India. P. Indian A.S. Plant S.C. 93:119–133.

State of Florida. 2001. Rules of the Department of Agriculture and Consumer Services, Division of Plant Industry, Chapter 5B-57, Introduction or Release of Plant Pests, Noxious Weeds, Arthropods and Biological Control Agents: Web page: http://doacs.state.fl.us/~pi/5b-57.htm. Accessed: December 15, 2003.

Strandberg, J. O. 1999. Pathogenticity of the fern anthracnose fungus, *Colletotrichum acutatum*, on wild and cultivated ferns in Florida. Proceedings of the Florida State Horticultural Society 112:274–277.

Stocker, R. K., A. Ferriter, D. Thayer, M. Rock, and S. Smith. 1997. Old World climbing fern hitting south Florida below the belt. Wildland Weeds 1:6–10.

Sundell, E. 1986. Noteworthy vascular plants from Arkansas. Castanea 51:211–215.

Sugai, M., K. Takeno, and M. Furuya. 1977. Diverse responses of spores in the light-dependent germination of Lygodium japonicum. Plant Sci. Letters 8:333–338.

Takeno, K., and M. Furuya. 1980. Sexual differentiation in population of prothallia in *Lygodium japonicum*. Bot. Mag. Tokyo 93:67–76.

Taylor, E. L. and C. D. Foster. 2003 . Managing your East Texas forest for the production of pine straw, Texas Cooperative Extension Service Publication 805-113. College Station, TX: Texas A&M University. 10 p.

Tranel, P. J. and T. R. Wright. 2002. Resistance of weeds to ALS-inhibiting herbicides: what have we learned? (Review) Weed Sci. 50:700–712.

Tu, M., C. Hurd and J. M. Randall. 2001. Weed Control Methods Handbook: The Nature Conservancy: Web page: http://tnc.ucdavis.edu. Accessed: May 20, 2006.

[USDA] U. S. Department of Agriculture. 1980. Soil Survey of Santa Rosa County, Florida. USDA Soil Conservation Service. 134 p.

[USDA] U. S. Department of Agriculture. 2006a. Germplasm Resources Information Network, *Lygodium japonicum*: Web page: http://www.ars-grin.gov/cgi-bin/npgs/html/taxon.pl?403435. Accessed: May 28, 2006.

[USDA] U. S. Department of Agriculture. 2006b. The PLANTS Database, National Plant Data Center, *Lygodium japonicum*: Web page: http://plants.usda.gov. Accessed: June 7, 2006.

University of Mississippi. 2006. Pullen Herbarium Specimen Database: Web page: http://herbarium.olemiss.edu/projects.html. Accessed: June 7, 2006.

University of North Carolina. 2006. The University of North Carolina Herbarium, Distribution maps: Web page: http://www.herbarium.unc.edu/distribution.htm. Accessed: June 7, 2006.

University of South Carolina. 2006. South Carolina Plant Atlas: Web page: http://cricket.biol.sc.edu/herb/. Accessed: June 6, 2006.

Valdosta State University. 2006. The Valdosta State University Herbarium Accession Database, *Lygodium japonicum.* Valdosta, GA: Valdosta State University.

Valenta, J. T., M. Zeller, and A. Leslie. 2001. Glyphosate controls Japanese climbing fern in experimental plots (Florida). Ecological Restoration 19: 118–119.

Van Loan, A. 2001. Weeds in the Woods. Florida Forests, (Fall 2001):27–29.

Van Loan, A. N. 2006. Japanese climbing fern: the insidious "other" *Lygodium*. Wildland Weeds 9:25–27.

Vitousek, P. M., C. M. D'Antonio, L. L. Loope, and R. Westbrooks. 1996. Biological invasions as global environmental change. Am. Sci. 84:468–478.

Volin, J. C., M. S. Lott, J. D. Muss, and D. Owen. 2004. Predicting rapid invasion of the Florida Everglades by Old World climbing fern (*Lygodium microphyllum*). Divers. Distrib. 10:439–446.

With, K. A. 2002. The landscape ecology of invasive spread. Conserv. Biol. 16:1192–1203.

Wilson, K. A. 2002. Continued pteridophyte invasion of Hawaii. Am. Fern J. 92:179–183.

Wunderlin, R. P. 1998. Guide to the Vascular Plants of Florida. Gainesville, FL: University Presses of Florida 806 p. .

Wunderlin, R. P., and B. F. Hansen. 2003. Atlas of Florida Vascular Plants, Florida Center for Community Design and Research, Institute for Systematic Botany, University of South Florida, Tampa: Web page: http://www.plantatlas.usf.edu/. Accessed: December 1, 2005.

Wynne, G. M., L. N. Mander, N. Goto, H. Yamane, and T. Omori. 1998. Biosyntheic origin of the antheridiogen, gibberellin A73 methyl ester, in ferns of the *Lygodium* genus. Tetrahedron Lett. 39:3877–3880.

Zeller, M. and D. Leslie. 2004. Japanese climbing fern controls in planted pine. Wildland Weeds 7:6–9.

Zimmerman, D. W. 1998. Invalidation of parametric and nonparametric statistical tests by concurrent violation of two assumptions. J. of Experiment. Ed. 67:55–68.

BIOGRAPHICAL SKETCH

Andrea (Andi) Van Loan has gained her greatest inspiration in life from the natural world, fostered and supported by her mother's appreciation and love of the same. Andi graduated from the Illinois Mathematics and Science Academy in Aurora, Illinois in 1990. After completing her Bachelor's degree in Resource Ecology and Management at the University of Michigan's School of Natural Resources and the Environment in 1994, Andi spent a brief time as a forester in Texas before moving to Florida. Andi has worked for the Florida Division of Forestry since 1995, first building the Ecology Unit at the Withlacoochee State Forest, and then advancing the agency's statewide focus on non-native plant species through work as a biologist in the Forest Health Section. Andi enjoys time spent with her family, including: husband, Chris Van Loan, father Harry Christman, brother Eric Christman, sister-in-law Connie Christman, and nephews Connor and Strider Christman. Andi's mother, Barbara Christman, will always serve as her inspiration in love of the natural world, and the barometer by which Andi's capacity to treat other people fairly and with open caring will be measured.