

COLUBRINA ASIATICA (LATHER LEAF) MANAGEMENT PLAN

COLUBRINA TASK FORCE

FLORIDA EXOTIC PEST PLANT COUNCIL



COMPILED BY:

Cheryl M. McCormick¹

PRINTED BY:

South Florida Water
Management District
West Palm Beach, FL

January 2007

Cover drawing courtesy Center for Aquatic and Invasive Species, University of Florida

¹ Former address: Center for Aquatic and Invasive Plants, University of Florida, Gainesville, Florida, 32653. Current Address: Santa Lucia Conservancy, 27600 Rancho San Carlos Road, Carmel, California, 93940; (831) 626-8595; cmccormick@slconservancy.org

Acknowledgements

The author would like to express appreciation and gratitude to Ken Langeland, Center for Aquatic and Invasive Plants, University of Florida, Tony Pernas, National Park Service, Florida and Caribbean Office, Jonathan Taylor, Everglades National Park, Terry Doyle, Ten Thousand Islands National Wildlife Refuge, Jim Duquesnel, John Pennecamp State Park, Shelby Moneysmith, Biscayne National Park, Terry Doyle, Ten Thousands Islands National Wildlife Refuge, and Pam Keyes, Rookery Bay National Estuarine Research Reserve for their time and cooperation in providing information related to case study development. David Jones, currently with Florida Atlantic University, inspired me both with his passion for Everglades National Park and contempt for latherleaf and all other biological invaders that threaten the Park's biological integrity. Finally, I would like to extend my thanks to the FLEPPC Board of Directors for being supportive of, and enthusiastic about, efforts directed towards eliminating non-native invasive plants from Florida's natural areas. Their enthusiasm and determination are exemplary... and infectious.

Colubrina Task Force Contact Information

Cheryl McCormick, Chair
Center for Aquatic and Invasive Plants
Institute of Food and Agricultural Sciences
University of Florida
7922 NW 71st Street
Gainesville, Florida 32653
cheryl@ufl.edu
(352) 846-2516 (O)
(352) 392-3462 (F)

The First Edition of the Lather leaf Management Plan compiled by the *Colubrina* Task Force provides comprehensive information, references, and contacts to aid in the development of integrated management strategies for eliminating established populations of *Colubrina asiatica* (L.) Brongn. (lather leaf) in coastal south Florida.

It is a compilation and survey of all known pertinent domestic and overseas literature describing the species in its native and host range to the date of publication. The Plan will be updated every five years, or as needed, to reflect changes in the invasive species literature, control techniques, and management philosophies concerning this species.

It is expected that not all of the information contained in the Plan will be applicable to all management scenarios. In some cases, the information contained herein may need to be adapted to local situations, issues, and problems.

Mention of trade names or proprietary product names does not constitute a guarantee or warranty of the product by the lather leaf Task Force or the Florida Exotic Pest Plant Council. There is no expressed or implied warranty as to the fitness of any product discussed. Any product name discussed is listed solely for the benefit of the reader, and the list may not contain all products available due to fluctuations in market conditions.

TABLE OF CONTENTS

I. INTRODUCTION	1
Problem Statement	5
Objectives	7
Recommendations	8
II. SPECIES BACKGROUND	9
Taxonomy	9
Description	10
Distribution	12
Native Countries of Origin	12
Host Range	13
Ethnobotanical Uses	13
Secondary Metabolites	14
Ecological Significance in Native Range	15
Impacts to Vegetation Communities	17
Competitive Mechanisms of Invasion	18
Reproductive Biology	19
Pollen Viability	19
Flower and Fruit Development	20
Seed Ecology	21
Germination Success	21
Greenhouse Experiments	21

In-Situ Experiments	22
Viability of Seed	23
Light Requirements for Germination and Establishment	24
Soil Seed Bank Density, Diversity, and Persistence	25
Dispersal	26
Impacts of and Recovery from Hurricanes	27
Seed embryo mortality	27
Stand Recovery relative to Native Species	27
Allelopathy	30
Impacts to Cultural Resources	31
Mapping Efforts	32
III. MANAGEMENT	
Regulatory Status	34
Chemical Control	35
Mechanical Control	35
Natural Enemies	36
IV. CASE STUDIES	37
Everglades National Park	37
John Pennecamp Coral Reef State Park	40
Biscayne National Park	42
Ten Thousand Islands National Wildlife Refuge	45
REFERENCES	48

Introduction

The establishment of some non-native plant species may pose a significant threat to native species diversity, ecosystem structure and function, human health, and the economy (Lodge *et al.* 2006; Pimental *et al.* 2000). Overall, only a fraction of introduced non-native plant species become established, viable populations, and a smaller fraction of these will emerge as direct threats to ecosystem functions or pose economic burdens (Williamson, 1996). Nevertheless, the number of invasive plant species in the U.S. remains formidable and continues to grow due to increasing global travel by humans and transportation of goods, increasing the need for early detection of and rapid response to, new invaders (Lodge *et al.* 1996; Vitousek *et al.* 1997; Crall *et al.* 2006).

The metamorphosis from immigrant to invader is a slow process – often cheekily referred to as an “explosion in slow motion”. A number of pernicious plant invaders experience initially slow rate of range expansion in their introduced range (a “lag” phase), which may be indistinguishable from the growth rates of non-invasive species. The lag phase is followed by a rapid exponential rate of proliferation and range expansion (a “log phase”) until the species reaches the geographic/physiological extent of its host range, and its population growth rate begins to level off (Mack *et al.* 2000) (Figure 1).

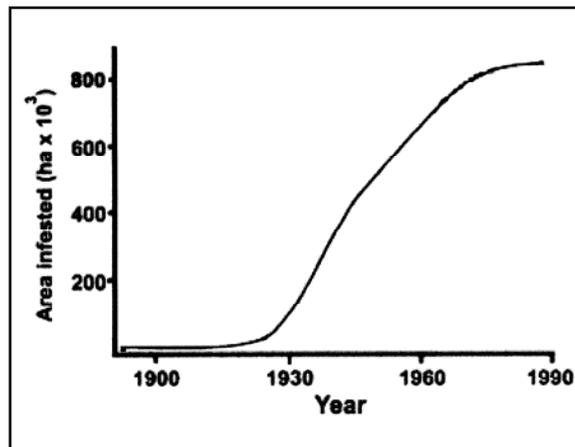


Figure 1. Plant invaders often exhibit exponential patterns of growth in their host ranges following extended periods of a “lag phase”, during which time growth and population expansion may remain undetected (from Mack *et al.* 2000).

In the case of Chinese tallow (*Sapium sebiferum*), an invasive tree of southern wetlands, the lag phase of population growth spanned almost a century in some parts of its range before there was evidence to suggest that the species was capable of modifying ecosystem structure and function. The lag phase of slow growth and minimal impacts represents the ideal “window of opportunity” at which early detection and prevention *should* (but rarely actually *does*) occur. Identifying potential invaders before they pose a problem and performing thorough risk assessments and is a major challenge for ecologists, policy makers, and land managers, and is partially attributed to the fact that invasive species management is, essentially, a *crisis management* field. Resources for invasive species management, including time, funding, manpower, and institutional/individual knowledge, are *always* limited - agencies and institutions seldom allocates adequate funding for non-crisis situations. Under these circumstances, we prioritize our invasive plant issues, take stock of existing resources, and practice triage rather than prevention.

What framework should drive research and management priorities for invasive plant species? Sustainable, long-term control strategies should be based upon ecosystem properties, particularly ecosystem vulnerability and conservation/production value (Hobbs and Humphries, 1995). This strategy shifts focus away from the ubiquitous species-by-species management paradigm that currently dominates the field, and a departure from triage, and towards healthy ecosystem structure and function, in which the invading plant species is a single component. It has become clear that ecosystems differ in their vulnerability to invasion and that the degree of susceptibility can be modified through prudent management practices (Crawley, 1987; Stohlgren *et al.*, 2004). For example, it is well-established that disturbance is an important factor influencing invasion dynamics (Rejmànek, 1989). The term ‘disturbance’ may be defined in terms of natural occurrences, such as floods and storm events, or human-induced, such as soil tillage and modified fire regimes. Not all disturbance regimes enhance the probability of invasion, and the type, intensity, distribution, and size of the disturbance should also be considered. Humphries *et al.* (1991) have asserted that attempts to manage invasive plant species without consideration of ecosystem properties are destined for failure, because they treat the symptoms, rather than the

cause of, invasion. Modifications to causal ecosystem linkages that facilitate the process of invasion must be addressed if sustainable, long-term, effective weed management will be realized.

Research on the Everglades “hole-in-the-donut” provides a poignant example of the need to identify causal mechanisms prior to initiating a costly, resource-intensive, large-scale management plan. In an effort to control an aggressive invasion of Brazilian pepper (*Schinus terebinthifolius*), Doren *et al.* (1991) determined that manipulating prescribed fire regimes had no significant effects on BP population density or growth. Instead, they determined that BP invasion was facilitated primarily by agricultural practices during the 50’s through the 70’s. Rock ploughing by farmers pulverized the limestone substrate and significantly altered both hydrologic and nutrient regimes. This massive alteration to ecosystem structure and function presented an insurmountable obstacle to “routine” management strategies. To counter this, the 9900-acre “hole-in-the-donut” project (a fraction of the total infested area) has cost over \$100 million to date, and was achieved by a massive effort involving scraping the soil to bedrock, hauling the soil off-site for incineration, followed by opportunistic colonization of pioneer species. An additional factor to consider in ecosystem management is post-eradication dynamics, particularly when the removal of one invasive species results in the growth of another.

An unforeseen complication of the “hole-in-the-donut” restoration effort has been a shift in food-web dynamics, whereby the prey-base for the now-infamous Burmese python, namely marsh rabbits, has grown significantly. NPS staff biologist Ray (“Skip”) Snow cannot state with certainty whether or not local populations of pythons have escalated in response to the increased resource.

As conservationists, resource managers, and policy makers, we need to adopt a long-term sustainable strategy for dealing with non-native invasive plant species in Florida’s natural areas. Current management paradigms are ecologically simplistic and do not address casual mechanisms of invasion. Across agencies, municipalities, and institutions, we need to adopt an integrated program of early detection and prevention, early control, and ecosystem management implemented at all stages of invasion (introduction, establishment, spread, and impact) that elucidate causal linkages

between disturbance regimes and invasion process (Lodge *et al.*, 2006; Hobbs and Humphries, 1995).

Biological invasions are significantly modifying Florida's native flora and faunal communities and their unique properties. If we are not vigilant and fail to implement effective management strategies to thwart the impacts of our most aggressive plant invaders, we risk losing the very character of Florida's natural resources.

Problem Statement

Lather leaf, *Colubrina asiatica* Brongn. (Rhamnaceae) is an invasive scandent shrub that has become naturalized in frost-free areas of the coastal southern peninsula and the Florida Keys (Godfrey and Wooten, 1981). This perennial shrub of pan-tropical origin and distribution was first documented in Florida Keys in the late 1930's, and most likely arrived there as a result of ocean currents and storm tides (Russell *et al.* 1982; Carlquist, 1974). From remotely sensed data and manual mapping efforts, we know that *Colubrina* is rapidly expanding its range in south Florida. Jones (1997) reported that, based on records and map products from the 1970's, *Colubrina* was capable of doubling its geographic range every 8 to 10 years.

Lather leaf invades coastal tropical hardwood and buttonwood hammocks, mangrove swamps, beach dunes, coastal strands, and tidal marshes (Myers and Ewel, 1990; Schultz, 1992; Jones, 1996). It is frequently found as impenetrable thickets growing at the interface of upland and submerged habitats, or between beach dune and maritime hammocks (Schultz, 1992). Like many pernicious woody invaders such as melaleuca (*Melaleuca quinquenervia*), Brazilian pepper (*Schinus terebinthifolius*), and Australian pine (*Casuarina equisetifolia*), lather leaf appears to be competitively superior in its host range, has no generalist predators or pathogens, and may be capable of significantly altering ecosystem structure and function in a relatively short period of time.

Observational and anecdotal information notwithstanding, very little empirical data has been published regarding the basic ecology of this potentially pernicious species, to say nothing of possible invasion mechanisms it may employ to successfully establish, compete, and produce viable populations in its invaded range. Recent research investigating various aspect of seed ecology of the species has begun, but much remains to be done to address the gaps in our collective knowledge base for lather leaf.

The lather leaf Management Plan has been developed to: 1) review the relevant literature pertaining to this aggressive species; 2) develop a framework for agencies mandated to protect Florida's natural areas from invasion by lather leaf; 3) to inform

managers of the most effective control techniques, and; 4) to identify gaps in knowledge with regard to this species, such that future research objectives may be clearly directed.

Goal Statement

The overall goal of the *Colubrina* Task Force is to develop a state-wide plan to protect and preserve the biological integrity of Florida's natural areas from deterioration by *Colubrina* invasion.

Objectives

A significant decrease in the distribution of *Colubrina* populations in Florida may be accomplished by establishing consistent, long-term management strategies that incorporate the following objectives:

1. Develop and implement best management practices (BMP's) based on integrated pest management (IPM) techniques addressing the causal mechanisms of ecosystem invasion.
2. Initiate a public outreach and education program to disseminate information regarding the ecology, invasion mechanisms, and environmental/economic impacts of *Colubrina* infestation in coastal south Florida.
3. Provide leadership and guidance in coordinating training opportunities through demonstration projects featuring integrated strategies for controlling *Colubrina* on public lands and cosmopolitan areas.
4. Coordinate efforts to secure support and resources, such as funds, labor, logistics, and knowledge from the Exotic Pest Plant Councils (EPPCs), federal (e.g., National Park Service, Fish and Wildlife Service) and state agencies (e.g., Water Management Districts, Department of Environmental Protection), NGO's (e.g., The Nature Conservancy), academia (e.g., The University of Florida), and international institutions (e.g., CABI Bioscience Centre, Switzerland) for the integrated management and control of *Colubrina* in Florida.

Recommendations

- 1. Adopt an ecosystem management approach:** Investigate the role of natural and anthropogenic disturbance in increasing ecosystem vulnerability to invasion by *Colubrina* and prioritize management plans to address causal mechanisms of modification to ecosystem structure and function by aggressive *Colubrina* infestation.
- 2. Encourage early detection and rapid response:** The most cost-effective, long-term strategy to decrease the spread of *Colubrina* populations in Florida is more natural resource managers to develop a rapid response system to detect and eradication small nascent populations before they become untenable. Currently, limited staff and remoteness of *Colubrina* populations (most are accessible only by boat) pose a challenge to detection.
- 3. Address gaps in knowledge and research:** Fund and support research addressing basic ecology and optimal management strategies of *Colubrina*
- 4. Partner with various stakeholders** (federal and state agencies, NGO's, academia, etc.) to produce public outreach and education materials for distribution to the general public to increase awareness about the environmental and economic impacts of *Colubrina* on public lands.
- 5. Increase the knowledge base of land managers:** Natural resource managers should be well versed in the correct identification and existence of *Colubrina* infestations on their lands, the basic ecological characteristics of the species, and the most effective control strategies for managing *Colubrina* populations in public lands.

Taxonomy

Colubrina asiatica (L.) Brongn.

Common Names: *Colubrina* (USA), Asian nakedwood, Asian snakewood, hoop wither (Caribbean), msuko (North Zanzibar), anapanapa (Hawaii), tartarmoana, soap bush (Republic of Fiji), kaka kaka (Papua New Guinea), beach berry bush (Australia), Tunhiriya (Sri Lanka), tutu (Tahiti), fîsoa (Samoa), kabatiti (Philippines), hanoh (Gambia)

Synonyms for this species include the following (Wunderlin and Hansen, 2002):

<i>Ceanothus asiaticus</i> L.	<i>Rhamnus asiatica</i> (L.) Lam. ex Poir.
<i>Ceanothus capsularis</i> (G. Forst.)	<i>Rhamnus splendens</i> Blume
<i>Celastrus sepiarius</i> Dennst.	<i>Sageretia splendens</i> (Blume) G. Don

<i>Pomderria capsularis</i> (G. Forst.)	<i>Tubanthera kataoa</i> Raf.
---	-------------------------------

Kingdom:	Plantae
Subkingdom:	Tracheobionta
Superdivision:	Spermatophyta
Division:	Magnoliophyta
Class:	Magnoliopsida
Subclass:	Rosidae
Order:	Rhamnales
Family:	Rhamnaceae
Genus:	<i>Colubrina</i>
Species:	<i>asiatica</i>

Related species in Florida (from Coile, 2001):

- C. arborescens* (Mill.) Sarg., - Coffee *Colubrina* – endangered
- C. cubensis* (Jacq.) - Cuban nakedwood – endangered
- C. elliptica* (Sw.) Briz. & Stern - Nakedwood - endangered



Figure 2. Native *Colubrina* species in south Florida. Both *C. arborescens* (coffee colubrine) and *C. elliptica* (soldierwood) are found in tropical hardwood hammocks in southern Everglades and adjacent areas (Armentano *et al.*, 2003). All three species have been placed on the state endangered species list (Images courtesy of Keith Bradley).

Description

The genus *Colubrina* is floristically the least specialized members of the Rhamnaceae (Buckthorn) family (Johnston, 1971). Within the U.S., approximately 20 species exist, of which three are native to Florida (Figures 2 and 3). Initially described by Alexandre Brongniart in 1826, *C. asiatica* (L.) Brongn. (common name: lather leaf) is a glabrous, evergreen, scandent or sprawling shrub capable of attaining heights of up to 10 meters; branches reddish-brown, slender, diffuse or prostrate (rarely erect and tree-like), up to 5 m in mature plants; branchlets are slender and may slightly “zig-zag”, with internodes from 5 to 50 mm long. Bark is dark brown, often with white striations. Leaves alternate, ovate, finely crenate-serrulate with 3-5 conspicuous primary veins and several pairs of lateral veins; petioles slender, to 10-15 mm long. Conspicuous dark green and shiny above and dull, paler green below (Johnston, 1971). Flowers reduced, cream-to yellow-green in color, approximately 4 mm in length, and borne in clusters at leaf axils; each with a nectar disc, 5 sepals, 5 hooded petals, and 5 stamens (Langeland and Craddock Burks, 1998; Jones, 1996). Medan and Hilger, 1992). Flowers are protandrous, rarely more than two of each cyme producing fruit, the oldest, lowest ones usually fertilized and then the upper ones often abortive, or if the early ones do not set fruit the later ones do; sometimes the upper flowers appearing unisexual (staminate) (Medan and Hilger, 1992). Jones (1996) reports that in southern Florida, flowering occurs in July. However, Long and Lakela (1971), Wunderlin (1982) and McCormick (*pers. observation*) have documented year-round flowering and fruit-set. Fruits a small (approximately 8 mm wide), tri-carpellate, subglobose capsule, fleshy and green when immature, becoming brownish-red upon ripening (Figure 3). Seeds are small (4.5-6 mm long) and buoyant, with a thin, dull, brown seed coat; released via tardy to prompt, sometimes explosive dehiscence (Johnston, 1971; Weber, 2003; Medan and Hilger, 1992). Johnston (1971) describes the seed coat as “easily breached”, suggesting no coat-induced physical dormancy for the species.

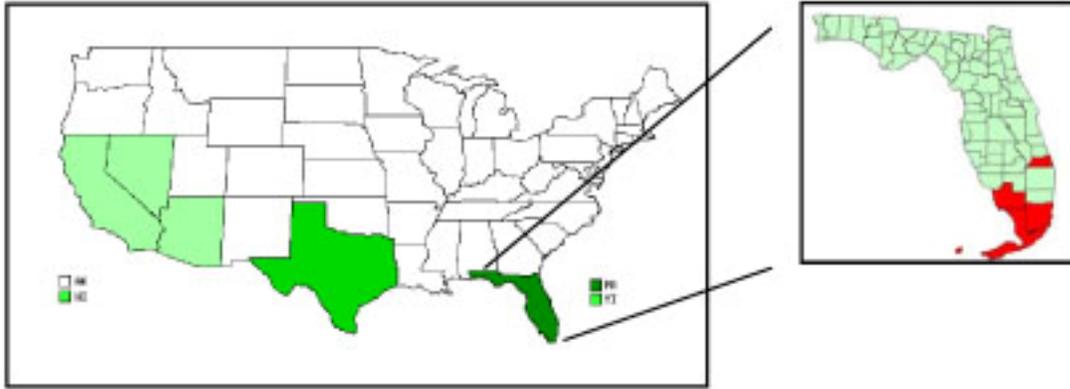


Figure 3. Distribution of the genus *Colubrina* in the conterminous U.S. and territories. Species present in Florida include: *C. arborescens*, *C. asiatica*, *C. cubensis*, and *C. cubensis* var. *floridana*. Source: Texas A&M, 1999 (<http://www.csd.tamu.edu/FLORA/>).



Figure 4. Its distinct morphological features, including scrambling growth habit, dark, shiny green leaves, small creamy- to greenish-yellow flowers, and dry, round fruits the size of a pea, makes *C. asiatica* relatively easy to identify in natural areas. Image of *C. asiatica* flowers used with permission, courtesy of CAIP-APIRS (APIRS photographs by Vic Ramey, 2000).

Distribution

Native Countries of Origin

The genus *Colubrina* is comprised of more than 30 species of trees and shrubs of nearly pan-tropical distribution, centered in tropical America, with a few species native to southeastern Asia, Malesia, tropical Australia and Polynesia (to Hawaii), with one species in coastal east Africa and the Mascarene Islands (Brizicky, 1964).

Colubrina has a vast pan-oceanic distribution, and is described by Johnston (1971) as being “native to...”strands and coastal lowland scrub of eastern Africa (Mozambique Kenya), Tanzania, Madagascar, the Seychelles, east and northeast to Ceylon and extreme southeastern India, southern Burma, Andaman and Nicobar Islands, Sumatra, Java, Borneo, the Malay Peninsula, Thailand, Taiwan, Cambodia, Vietnam, Hainan, Ryukyu Islands, Fiji Islands, Samoan Islands, New Hebrides (now Vanuatu), Tonga and Society Islands, New Caledonia, Cook Islands, Marquesas, Tuamotu, Papua New Guinea, Australia (extreme north Queensland) and Hawaii” (Figure 5).

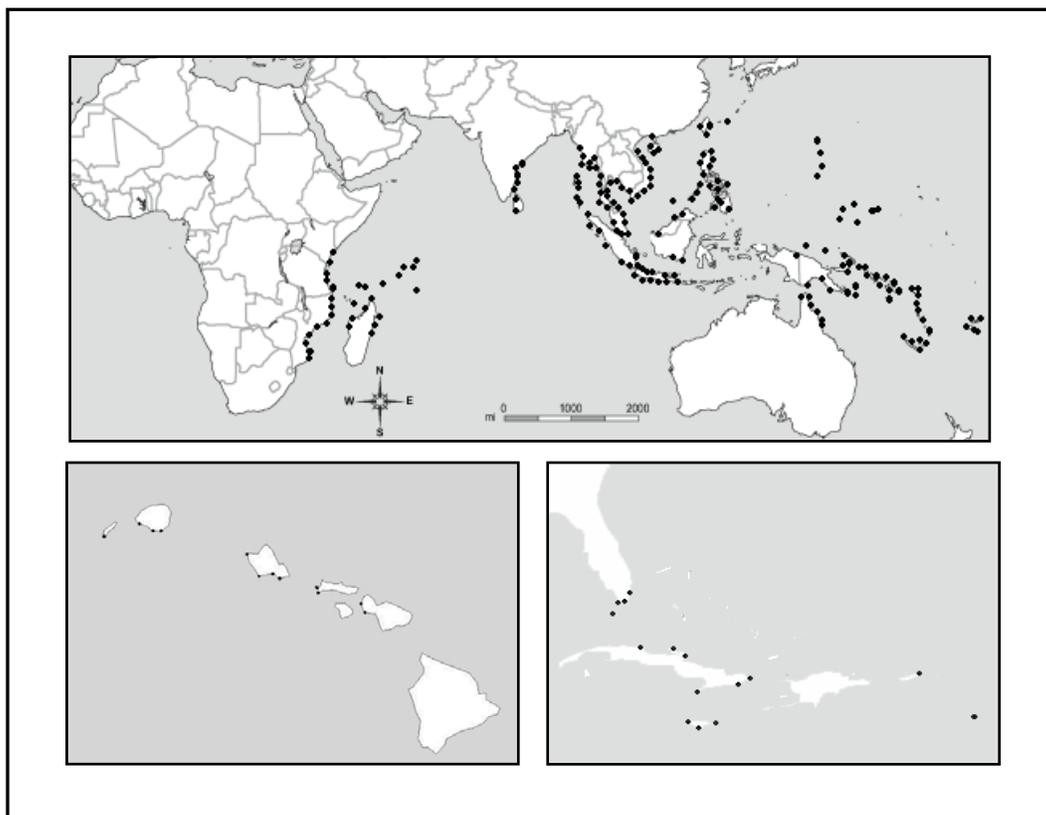


Figure 5. Pan-tropical Old World distribution of *C. asiatica* (top); distribution in the Hawaiian island chain (bottom left) and south Florida and the Caribbean (bottom right). Adapted from Johnston, 1971).

Host Range

Colubrina is a relatively recent introduction to the Western Hemisphere. The species was documented in Jamaica in the 1860's by the botanist March, and may have been introduced there by East Asian immigrants who used the plant in traditional medicine and as a soap substitute (Johnston, 1971). In his description of *Colubrina* in Jamaican habitats, Adams (1972) notes that the species is "common in coastal thickets and on sandy and rocky shores and cays". Viable seed is believed to have been dispersed via thalassocchory (ocean currents) to other islands within the Caribbean Basin, such as the Bahama Archipelago, Grand Cayman, Cuba, the Dominican Republic, Jamaica, Haiti, Martinique, Mexico, and south Florida, where it has become naturalized" (Johnston, 1971; MOBOT, 2006). On Grand Cayman Island, *Colubrina* appears to have become more abundant following Hurricanes Andrew (1992) and Ivan (2004). Ann Stafford, a veteran naturalist with the Grand Cayman National Trust describes her observations of *Colubrina* in the Districts of the Island, noting that the "tangled thickets made (the) dyke road impassable".

The first recorded voucher specimen of *Colubrina* in North America was collected by Killup in 1937 from Big Pine Key. The species was observed by biologists in north Key Largo in 1951, in Key West in 1963, and on Key Biscayne in 1966 (Russell *et al.*, 1982). Specimens were first collected in Everglades National Park by Craighead in 1954 on Crocodile Point Road, and in 1961 at Flamingo (Russell *et al.*, 1982). *Colubrina* has been reported in scrub and/or coastal areas, beaches, back dune habitats and tropical hardwood hammock margins from Key West north to Hutchinson Island (St. Lucie County), in Everglades National Park, including the Ten Thousand Islands northwest into Collier County (FLEPPC, 1996; Langeland and Craddock Burks, 1998), and as far east as Elliott Key in Biscayne National Park, Dade County.

Ethnobotanical Uses

As its common name implies, the leaves of *Colubrina*, when crushed, possess lathering properties (Johnston, 1971). In the In Samoan and the Fijian Islands, the leaves are crushed and used as a detergent and shampoo ("Vuso levu"). The author

has found that fresh leaves, when placed in a blender and crushed with water, produces copious amounts of *foam*, but observed no detergent or soap-like properties associated with the frothy mixture. Perry and Metzger (1980) report that, when macerated, the resulting mixture of saliva-macerated leaf mixture is applied to burns caused by centipede or millipedes (Petelot, 1952). In the Bahamas, *Colubrina* is/was used as a digestive aid, antiscorbutic (counteracts scurvy), tonic, laxative, a febrifuge (reduces fever), medicinal bath, and a vermifuge (expels intestinal parasites) (Austin, 1999; Morton, 1981; Burkill, 1966). In the Philippines, an extract of the leaves is used to lessen topical irritation and to remedy skin diseases (Guerrero, 1921). A distilled mixture of whole fruits is reportedly used as an abortifacient as well as a piscicide and migraine remedy (Uphoff, 2001; Quisumbing, 1951; Gimlette, 1929; Guerrero, 1921). In Sri Lanka, a cottage industry has developed from cutting and drying *Colubrina* (colloquially called *Tunhiriya*) stems and weaving them into mats (Nikapitiya, 2005). The wood is dense with a distinct reddish-orange color, and when finished is used for small-scale woodworking adornment items, such as pool cues and knife handles (www.smartsnooker.com). In southern Thailand, it is cooked atop steamed fish, though it is not considered an economically important food source, *per se* (Paddle Asia, 2006).

Secondary Metabolites

Essential oil isolated from the seed kernel of *Colubrina* contains terpenes and other unsaturated compounds and are responsible for inhibiting the growth of a number of pathogenic microbes. Seed kernel oil exhibits “maximum inhibitory activity” against a number of pathogenic and non-pathogenic bacteriae, including *C. diptheriae*, *V. cholerae*, and *S. aureus* (Kar and Jain, 1971). Kar *et al.* (1970) report that the essential oil of the fruit has a blood pressure reducing effect and induces spasmolytic (seizure-inhibiting) activity when administered to rats. Isolated saponins show antagonistic effects on amphetamine and has shown to have sedative effects in mice (Wagner *et al.*, 1983). The leaves and bark contains various saponins, primarily Colubrin and Colubrinoside, as well as alkaloids (Colard *et al.*, 1976; Lee *et al.*, 2000) which, when isolated, produce the lathering properties for which the species is known. The leaves

also contain jujubogenin glycosides and flavinoids, which have been shown to exhibit antifungal properties (Hounsel, 2001). From the seed kernel, Tolkavech *et al.* (1980) isolated bisbenzylisoquinoline alkaloids and 2-benzylisouinoline compounds, which possess anti-hypertensive and anti-arrhythmic effects in hypertensive mice and humans. It is interesting to note that, although significant cancer inhibiting compounds have been identified and isolated in six New World species of the Genus *Colubrina*, no anti-cancer compounds have been isolated in *C. asiatica* specifically, to date.

Ecological Significance in Native Range

Often the best predictor of how an invasive plant species will behave in its introduced range is to investigate its ecology within the native range. Information regarding non-commercial species (i.e., those not utilized for forestry, agriculture, and/or horticulture) may be difficult to locate, but can be made more tenable by conducting internet searches within the host domains of each country to which a species is native. Additionally, natural resource managers, ecologists, and herbarium staff associated with native counties are usually eager to share information and resources.

Colubrina appears to possess a broad ecological amplitude throughout its native range. In a floristic survey of understory vegetation along the Wapoga River corridor in Irian Jaya (Indonesia), Mogeia (2000) noted that *Colubrina* was collected at an altitude of approximately 2300 to 2500 meters and was covered “from ground to tree branches” with moss (along with other species). This suggests that *Colubrina* is capable of establishing populations in temperate, moist, freshwater riparian habitats. On the small, mangrove-dominated island of Nasoata, Viti Levu (Fiji), *Colubrina* is found in association with littoral forest and strand vegetation. Soils subtending these communities are a mixture of terrestrial carried via river outflow, mixed with sand from coral reef and oceanic sources, and therefore high(er) in nutrient content than the sands of most littoral beaches (Thaman *et al.*, 2005). Nasoata is characterized by a mean sea level of 1.2 meters, an average annual temperature of 25°C (77° F), and a mean annual rainfall of 3,000 mm. Ghazanfar *et al.* (2001) observed that *Colubrina* was the dominant species on the seaward side of seven of nine islands surveyed off the eastern

and southeastern coasts of Viti Levu (Fiji). The islands have an oceanic climate and are heavily influenced by heavy salt spray, tsunamis, storm events, and cyclones, with an average rainfall of between 2,000 and 3,000 mm, and an average ambient air temperature of 24°C (75°F). Soils are composed of alluvium and beach sand.

On Venuatu (formerly New Hebrides), *Colubrinas* described as a common coastal strand species which "... provides the very important service of protecting gardens from salt spray and coastal erosion" (Clarke and Thaman, 1990). In their study examining successional vegetation replacing invasive *Chromolaena odorata* ("Jack in the bush", Asteraceae) after defoliation by *Pareuchaetes pseudoinsulata* (Lepidoptera: Arctiidae), a biocontrol agent, Marutani and Muniappan (1991) report that *Colubrina* was the dominant species one of three sites on Guam. Annual precipitation averaged 2338 mm, with a mean ambient temperature of approximately 25°C (77° F) and day length of between 11.3 (December) and 12.9 (June) hours. The Waimanalo Watershed Restoration Project of Aiea, Hawaii advocates using *Colubrina* ('Anapanapa') to control bank erosion along freshwater streams, stating that the species thrives in full sun to partial shade, medium- to wet soil conditions, and 0 to 300' in altitude (USDA-NRSC, 2006). In Queensland (Australia), *Colubrina* is primarily associated with dune and dune/swale communities, where it forms dense thickets along woodland/grasslands interfaces (TVE, 2006). In Samoa, the species is commonly associated with littoral scrub communities situated between littoral forest and herbaceous strand zones, and are subtended by sandy and/or rocky substrate (FAO, 2000). Although there is no evidence to suggest that *Colubrina* colonizes the understory of adjacent littoral forest communities in Samoa, in the Republic of Niue (approximately 460 km east of Tonga), *Colubrina* is noted as a common understory species of closed broadleaved littoral forests, and suggests that the species is capable of becoming established in low light environments. These habitats are subtended by shallow coralline substrate, and characterized by oceanic climates, high relative humidity, an average ambient temperature of 27°C (81°C) and elevation of approximately 61 meters above sea level. In the Philippines, 'Kabatiti', as it is colloquially known, is a common species on shorelines and along tidal streams, the latter subtended by highly organic, mucky soils. In his description of *Colubrina* in

Jamaican habitats, Adams (1972) notes that the species is “common in coastal thickets and on sandy and rocky shores and cays”.

Impacts to Native Vegetation Communities

Colubrina invades coastal ridges subtended by low permeability, marl soils, immediately above high tide line, in buttonwood, mangrove, and tropical hardwood hammocks and tidal marshes (Long and Lankela, 1971). Once established, *Colubrina* growth habit results in very dense, monotypic thickets up to several feet thick. *Colubrina* poses an immediate threat to the biodiversity, structure and function of habitats by outcompeting neighboring vegetation indirectly by exploiting shared resources more efficiently than native species, or directly by physically growing over subtending vegetation (interference competition) (Figure 6). Ecologically sensitive coastal communities in Everglades National Park that are especially vulnerable to *Colubrina* invasion include floristically unique tropical hardwood hammocks. These communities contain a number of Florida-listed threatened and endangered species, including West Indian mahogany (*Swietenia mahagoni*), thatchpalm (*Thrinax radiata*), wild cinnamon (*Canella winterana*), manchineel (*Hippomane mancinella*), cacti (*Cereus* spp.), bromeliads (*Tillandsia* spp.), and orchids (*Encyclia boothiana*, *Oncidium luridum*) (Jones, 1996).



Figure 6. *Colubrina* growing in the coastal ridge of Everglades National Park, where it grows atop native species and outcompetes neighboring vegetation for available resources. The understory is virtually impenetrable in a mature stand.

In the Florida Keys, *Colubrina* has been documented in every state park, and poses a threat to rare native species such as beach star (*Remirea maritima*) and bay cedar (*Suriana maritima*). There is no empirical or anecdotal evidence to suggest that *Colubrina* directly impacts endangered or threatened fauna. However, indirect impacts may be incurred to wildlife, particularly neotropical migratory avifauna, through habitat degradation, reduction in plant structure/richness/diversity, and outcompeting native species that provide food and/or refugia.

Competitive Mechanisms of Invasion

Colubrina may facilitate its own growth and spread in part by modifying light regimes under its canopy, such that the understory becomes characterized by very deep shade, rich in long-wavelength far-red light, to which coastal native species can neither readily use nor adapt to. In a year-long survey of *Colubrina* populations in south Florida, the author has observed no seedling recruitment from native dicot species in the understory of *Colubrina* thickets. Although *Colubrina* seedlings are observed, they are present in low densities (mean = $11 \pm 4.3 \text{ m}^2$) and tend to be thin-stemmed and etiolated. Etiolated (dark grown) seedlings have very high concentrations of phytochrome A, which make the seedling more sensitive to light, so that the seedling can be induced to produce its photosynthetic machinery when it encounters even very dim light. *Colubrina* seedlings may be adapted to early development in deep shade, followed by rapid growth once the overstory is opened or removed by disturbance (e.g., hurricane, herbicide treatment, fire, etc.) and seedlings are exposed to full or partial sunlight. Seeds receiving primarily far red light in nature are most likely surrounded by mature plants and are at a competitive disadvantage, so there is an adaptive advantage for these seeds not to germinate. Because in nature wind can temporarily move leaves allowing unfiltered sunlight to penetrate, seeds surrounded by other plants would occasionally receive unfiltered sunlight. The stimulatory effects of this light would have to be reversed by far red light when the wind dies down and the leaves of the surrounding plants once again shade the seeds, otherwise occasional sunflecks could induce the seeds to germinate even though they are still at a competitive disadvantage.

Ecophysiology studies investigating light quality and intensity in *Colubrina* understories may elucidate relationships between light dynamics, seed germination, and seedling growth and development.

Belowground interference competition for resources (i.e., space, nutrients, water), may be important for the species, and should be further investigated.

McCormick and Langeland (2007) observed that even seedlings had tremendous capacity for root growth (Figure 7). Neighboring species would presumably have to be superior competitors for underground resources, or develop novel strategies of parsing existing resources in ways sufficient for growth and reproduction.



Figure 7. *Colubrina* seedlings have potential to produce large root masses in a relatively short period of time. Competition for limited underground resources by roots may be a significant mechanism contributing to the success of *Colubrina*.

Reproductive Biology

Pollen Viability

Using aceto-carmin staining methods, Rigamoto and Tyagi (2002) investigated pollen viability for *Colubrina* and 31 additional coastal species on the remote Pacific

Island of Rotuma. Their results indicate that *Colubrina* (“tartarmoana”, as it is colloquially known) had a pollen fertility range of 38-49 percent (mean = 45 ± 3.2). Only three additional species tested showed pollen viability below 50%. Pollen fertility is a critical determinant of whether sufficient sexual reproduction will occur to ensure the survival of a species, and has been shown to be reduced in small, fragmented populations (Jennersten and Nilssen, 1993). Menges (1991) and Agren (1996) observed low pollen fertility and reduced seed germination rates in small, fragmented populations.

Pollen viability analysis has not been analyzed for *Colubrina* populations in south Florida. However, their spatial segregation, coupled with relatively low germination success, and extremely low incidence of insect pollinators on islands, suggests that *Colubrina* in south Florida reproduces primarily by asexual means, either via vegetative apomixis or agamospermy (Briggs and Walters, 1997).

Flower and Fruit Development

In their analysis of the reproductive structures of the members of the genus *Colubrina*, Medan and Hilger (1992) noted that *Colubrina* floral morphology and development are strongly indicative of protandry (the shedding of pollen of a plant or flower prior to receptivity on the same plant or flower) – a condition typical in the Rhamnaceae (Medan, 1991). In this fashion, plants prevent self-fertilization through temporal segregation of male/female floral structures. Additionally, the stamens of *Colubrina* flowers develop and move in a centrifugal fashion, providing additional protection against self pollen/stigma interference (Degener, 1946). Observations by Johnston (1971) that the upper flowers in the inflorescence appeared unisexual (staminate, i.e., male) was supported by Medan and Hilger. Johnston (1971) also noted that if basal (i.e., early) flowers fail to set fruit, the distal (i.e., later) flowers do, and suggests that functional specialization occurs within the inflorescence. During floral ontogeny, three pollen tube pathways develop independently and are linked by a single, basally-situated compitum (i.e., a tract of tissue down which pollen tubes grow). The significance of this floral configuration is not known, but Medan (1985) hypothesizes that, by providing a unique pathway for transport of pollen, excessive pollination tube

competition may be reduced, thereby ensuring optimal fertilization. Strong pollen competition, if coupled with poor pollinator visitation frequency, would lead to insufficient pollen tubes reaching the ovules (Medan and Hilger, 1992).

Seed Ecology

Seed is a critical stage of the plant life cycle and significant regulator of plant population dynamics. Paradoxically, seed ecology, dormancy, and germination traits of invasive species are rarely studied and poorly understood. A clear understanding of how *Colubrina* seed ecology influences population growth and how germination is regulated under natural conditions can enhance management of this species. By exploiting “windows of management opportunities” during which *Colubrina* populations may be particularly vulnerable, relatively small management initiatives may result in significant population regulation.

Germination Success

Greenhouse Experiments – Based on studies of seven populations in Everglades (including the lower Ten Thousand Islands) and Biscayne National Parks, McCormick and Langeland (2007) determined that germination success of *Colubrina* in 2005 ranged from 9.6% to 18.8%, and that rates varied significantly among populations ($p < .001$) (Figure 8a). The dataset for 2006, although not inclusive of all populations due to severe hurricane activity in 2005, shows a pronounced decrease in germination success (Figure 8b), and suggests significant annual and/or seasonal differences among populations. While these results were unexpectedly low, they are aligned with the findings of studies by other workers examining germination success of congeners in the Rhamnaceae family. For example, Obata (1967) found that untreated seeds of *C. oppositifolia* had germination rates ranging from 5 to 30%.

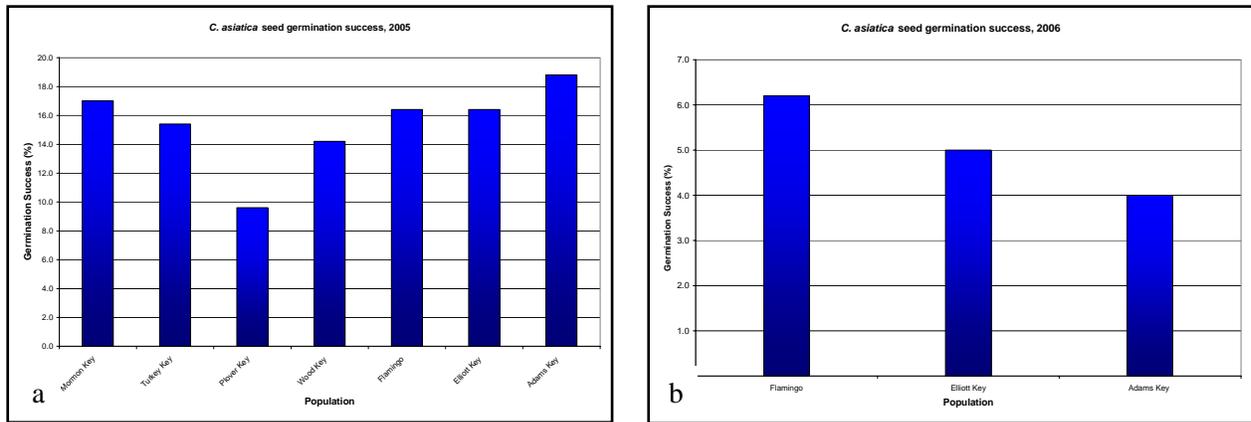


Figure 8. Seed germination results for field collections made in 2005 (a) and 2006 (b). Seeds from populations located in the lower Ten Thousand Islands were unavailable in 2006 due to severe hurricanes in 2005.

In-situ Experiments - To determine the effects of burial on *in-situ* germination, McCormick and Langeland (2007) conducted field experiments in which twenty *Colubrina* seeds were placed in each of thirty nylon mesh drawstring bags. Fifteen of the 30 bags were buried to a depth of 1 cm (~ 2.5") and the remaining bags were placed on the soil surface (Figure 9). Bags were examined once per month for a period of six months for evidence of successful germination, which was defined as emergence of radicle tissue. Of the 4,200 seeds (7 sites x 30 bags/site x 20 seeds/bag) analyzed in the study, only 34 seeds (0.81%) successfully germinated in the bags. However, germination did not occur for at least 4 months (Table 1). It is noteworthy that those seeds that did germinate did so in nylon bags that were buried; no seeds germinated from bags that placed on the soil surface without burial. Viability analysis conducted on the ungerminated fraction using standard Tetrazolium HCL ("TZ") testing (AOSA, 2000) revealed that 96% of embryos were aborted. Medan and Hilgar (1992), in describing the floral characteristics of the species, note that fruits produced in the uppermost portion of the cyme (i.e., a flat-topped flower cluster in which the main axis and each branch end in a flower that opens before the flowers below or to the side of it) are usually aborted. Culliney (1999) notes that some individual trees do not produce viable seeds at all - these seeds do not contain an embryo. These earlier findings are aligned with those of McCormick and Langeland (2007).



Figure 9. Nylon mesh seed bag attached to flagging used in *in-situ* soil seed burial experiments (left); *Colubrina* seeds germinating in nylon mesh bags after four months (center and right).

Table 1. Results of *in-situ* buried seed experiment over a period of six months. Germination events begin in the fourth month and remain relatively constant for the following two months.

POPULATION	30-DAYS	60-DAYS	90-DAYS	120-DAYS	150-DAYS	180-DAYS
Flamingo	0	0	0	0	0	0
Adams Key	0	0	0	0	3	3
Elliott Key	0	0	0	7	7	7
Wood Key	0	0	0	0	1	1
Plover Key	0	0	0	0	4	4
Turkey Key	0	0	0	0	7	7
Mormon Key	0	0	0	0	12	12

Viability of Seed

McCormick and Langeland (2007) tested the ungerminated fraction of 500 seeds of each population from which seeds were collected in the fall 2005 and used in germination studies. Additionally, seeds were collected from three populations in winter 2006 and analyzed. Four populations from the Ten Thousand Islands (i.e., Mormon, Turkey, Plover, and Wood keys) were excluded from the 2006 data set, because Hurricanes Rita and Wilma effectively eliminated or killed the existing seed crops of these stands.

The status of the embryo tissue of seed was determined using standard TZ staining techniques. The TZ is a biochemical test that differentiates live from dead seeds based on the activity of the respiration enzymes in seeds. Consistent with other seed traits for the species, there were no significant differences among populations in terms of embryo viability for either 2005 ($p = 0.1674$) or 2006 ($p = 0.1632$) seed crops.

Furthermore, seed viability for the species was extremely low for all populations and for both years (Table 2).

Table 2. Results of seed embryo viability analysis showing very low viability for *Colubrina* for seed crops produced in fall of 2005 and winter of 2006.

POPULATION	SAMPLE DATE	# SEEDS TESTED	VIABLE EMBRYOS	% VIABILITY
Mormon Key	10/06/2005	415	11	2.7
Turkey Key	10/12/2005	423	6	1.4
Plover Key	10/12/2005	452	14	3.1
Wood Key	10/05/2005	429	5	1.2
Flamingo	09/07/2005	418	20	4.8
Elliott Key	11/22/2005	418	5	1.2
Adams Key	11/21/2005	406	15	3.7
Flamingo	01/17/2006	1021	28	2.7
Adams Key	01/26/2006	1430	67	4.7
Elliott Key	03/07/2006	103	3	2.9

Light Requirements for Germination and Establishment

Jones (1996) states that the seeds require loose soil to germinate. McCormick and Langeland (2007) found that seeds germinated even when covered by moderately compacted sandy soil (i.e., beach sand). In contrast to reports by Schultz (1992) and Russell *et al.* (1982), which report that considerable light is required for germination and seedling growth, McCormick and Langeland (2007) observed seed germination and seedling growth in the understory of dense *Colubrina* thickets. However, such seedlings were etiolated and thin relative to seedlings grown in high light environments. When etiolated seedlings were transplanted into a high light environment (i.e., a greenhouse) they developed “normal” morphological features over time. It appears that, while the species certainly thrives in high-light environments, *Colubrina* may be capable of tolerating a very broad range of light conditions, and of altering microsites by creating deep shade conditions to which some conspecifics may tolerate but native species are not capable of rapidly evolving.

Soil Seed Bank Density, Diversity, and Persistence

McCormick and Langeland (2007) analyzed the contents of thirty, 12-cm³ soil samples from each of the seven sites in October-November 2005. Results indicate that all sites had low species richness and exhibited considerable overlap in species composition (Table 3). In general, soil seed bank densities were very low for all sites, regardless of species composition. Only two sites (Adams Key and Plover Key) had soil seed bank densities over 20 *Colubrina* seeds/12.5 cm³. In addition to whole, intact *Colubrina* seeds, there were many conspecific seeds in various states of decomposition and fungal decay, which was especially pronounced in sites underlain by soils containing high(er) amounts of organic content. Rapid seed decay was also observed during the course of exploratory greenhouse experiments in the spring of 2004, when the majority of *Colubrina* seeds buried for more than six months exhibited accelerated fungal decay and embryo death. These results suggest that the soil seed bank for *Colubrina* is transient (less than one year) rather than persistent (more than one year), and may be shorter for seeds in sites characterized by soils containing high levels of organic material.

Table 3. Species richness and abundance within and beyond stands of *Colubrina* for all sites.

SITE LOCATION	SPECIES WITHIN STAND	ABUNDANCE (COLUMN 2)	SPECIES BEYOND STAND	ABUNDANCE (COLUMN 4)
Flamingo	<i>Colubrina asiatica</i>	83	<i>Colubrina asiatica</i>	23
	<i>Poa</i> spp.	2	<i>Poa</i> spp.	1
Adams Key	<i>Colubrina asiatica</i>	115	<i>Colubrina asiatica</i>	75
	<i>Poa</i> spp.	5	---	---
	<i>Cinnamomum camphora</i>	2	<i>Cinnamomum camphora</i>	2
Elliott Key	<i>Colubrina asiatica</i>	17	<i>Colubrina asiatica</i>	2
	<i>Caesalpinia bonduc</i>	1	---	----
Wood Key	<i>Colubrina asiatica</i>	93	<i>Colubrina asiatica</i>	5
	<i>Rhizophora mangle</i>	4	<i>Rhizophora mangle</i>	5
Plover Key	<i>Colubrina asiatica</i>	110	<i>Colubrina asiatica</i>	0
	<i>Rhizophora mangle</i>	2	<i>Rhizophora mangle</i>	1
Turkey Key	<i>Colubrina asiatica</i>	11	<i>Colubrina asiatica</i>	9
	<i>Abrus precatorius</i>	3	<i>Abrus precatorius</i>	7
Mormon Key	<i>Colubrina asiatica</i>	43	<i>Colubrina asiatica</i>	2
	<i>Rhizophora mangle</i>	1	<i>Rhizophora mangle</i>	2

Dispersal

The seeds of *Colubrina* are morphologically and physiologically highly adapted to long-distance, oceanic dispersal (i.e., thalassochory). As noted by Guppy (1906) the seeds floated in sea water for many months without loss of viability. Guppy's observations were re-examined and confirmed by Carlquist (1966), who stated that "seeds of *Colubrina* float indefinitely, as Guppy claims."

Carlquist attributes the buoyancy of *Colubrina* seeds to a prominent space which forms between the cotyledons and the endosperm (Figure 10). If the testa is removed, the seed becomes permeable to water and subsequently sinks (McCormick, *pers. observation*). Explosive dehiscence accounts for proximate dispersal of propagules from the parent plant. Using scanning electron microscopy (SEM), Medan and Hilger (1992) observed that fracture lines appear to accumulate in the mechanical layers of the endocarp during fruit desiccation. These fracture lines lead to oblique bending and mutual repulsion, resulting in the "breaking" of tissue external to the endocarp.

Although some biologists speculate that birds may secondarily disperse the seeds to upland areas, there is no empirical or observational evidence to support the claim that birds utilize *Colubrina* as either a food source or a "crop stone". Given its adaptations for sea water dispersal, it does not appear to rely solely on "aboriginal introduction", as suggested by Brown (1935) to explain its wide distribution. Smith (1990) collected drift disseminule assemblages (seeds, viviparous seedlings, fruits, and their fragments) from nine beaches on and near Viti Levu, Fiji. He observed *Colubrina* in abundance, where it easily germinated on the beach strandline; he also noted that the seeds floated for more than one month.

... and found In a biogeographic floristic survey of southeast Polynesia, Kingston *et al.* (2003) noted that *Colubrinis* conspicuously absent from the Pitcairn Island group. The flora of the Pitcairn island group is a subset of the sub-set of the larger flora of southeast Polynesia (Austral, Cook, and Society Islands), but is out of the range of annual cyclones that affect these islands, which enhance dispersal between these island groups, suggesting that *Colubrinis* dispersed via episodic storm events. That stated, the species contains useful saponins and other secondary metabolites which may facilitate human introduction to some areas (see "ethnobotany").

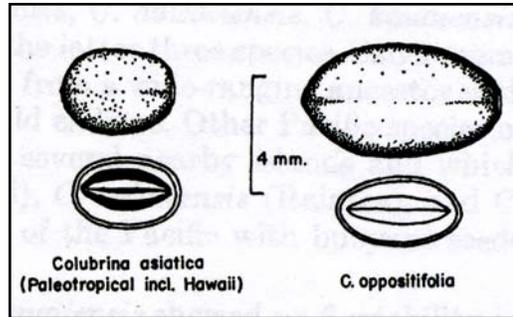


Figure 10. Seeds of endemic Hawaiian *Colubrina oppositifolia* compared with wide-ranging *C. asiatica*, which is dispersed by ocean currents. Prominent spaces between cotyledons and endosperm in *C. asiatica* is indicated in black (Carlquist, 1966; adapted from Hillebrand, 1888).

Impacts of and Recovery From Hurricanes

Seed embryo mortality – To determine what, if any, impacts hurricanes had on seed embryo mortality, McCormick and Langeland (2007) collected and analyzed *Colubrina* seeds from three populations in the lower Ten Thousand Islands before and after Hurricane Wilma. Of the 151 post-Wilma seeds tested for viability using TZ, no viable embryos existed; all were extremely desiccated and discolored. Additionally, seed coats were very brittle and golden yellow in color, as opposed to the normal phenotype which is a dull, dark gray-brown (Figure 11).

In addition, time-sequenced photographs were acquired from consistent locations (+/- 1-2 meters) to document *Colubrina* post-hurricane recovery and regrowth relative to native species over the course of 2006 (Figure 12). Based on these observations, it is evident that *Colubrina* is able to recover much earlier than natives following severe hurricanes. Not only was it the first species to vigorously flush with new growth following Wilma, but thickets appeared to thrive, perhaps in part due to the removal of dead biomass from the subcanopy, which allowed lower branches to resprout profusely with the influx of light, water, and nutrients. Additionally, mortality and/or removal of the existing seed crop allows plants to invest all available resources into growth, rather than allocating limited resources into the production of reproductive structures. Indeed, by August of 2006, stands of *Colubrina* appeared to be quite luxuriant relative to its storm-hardy, but still defoliated native neighbors, such as buttonwood (*Conocarpus erectus*), red mangrove (*Rhizophora mangle*) and sea grape (*Cocoloba uvifera*).



Figure 11. *Colubrina* seeds collected before (left) and after (right) Hurricane Wilma in the fall and winter of 2005. Hurricanes may contribute to high mortality of seed, and indeed may kill an entire seed crop through prolonged flooding.

The implications for such post-disturbance community dynamics are unknown. However, in its native range *Colubrina* exposed to cyclonic activity on a regular basis. Furthermore, the unspecialized seeds have been documented to be dispersed by ocean currents; *Colubrina* is well-adapted to the harsh environmental conditions that episodic, severe storm conditions create. In terms of south Florida, particularly in the Ten Thousand Islands and Everglades, winds and even severe flooding events may carry seed into sensitive habitat types, such as coastal hardwood hammocks, the species composition of which is unique in North America and is characterized by a number of endemic and rare species. Despite the general trend for the species (and perhaps, the Genus as a whole) to possess low germination success, short persistence in the soil seed bank, and low viability, seed crop production is massive, on a population level. Therefore, even a relatively low level of germination success can result in exponential growth of the species. Post-hurricane conditions, though potentially devastating to native species, may present an ideal window of opportunity for vegetation specialists to apply various treatments to *Colubrina*. Not only is it physiologically stressed, but also partially defoliated, at least for a period of time, and thicket density is greatly reduced, thus greatly enhancing physical access, visibility, and treatment success.

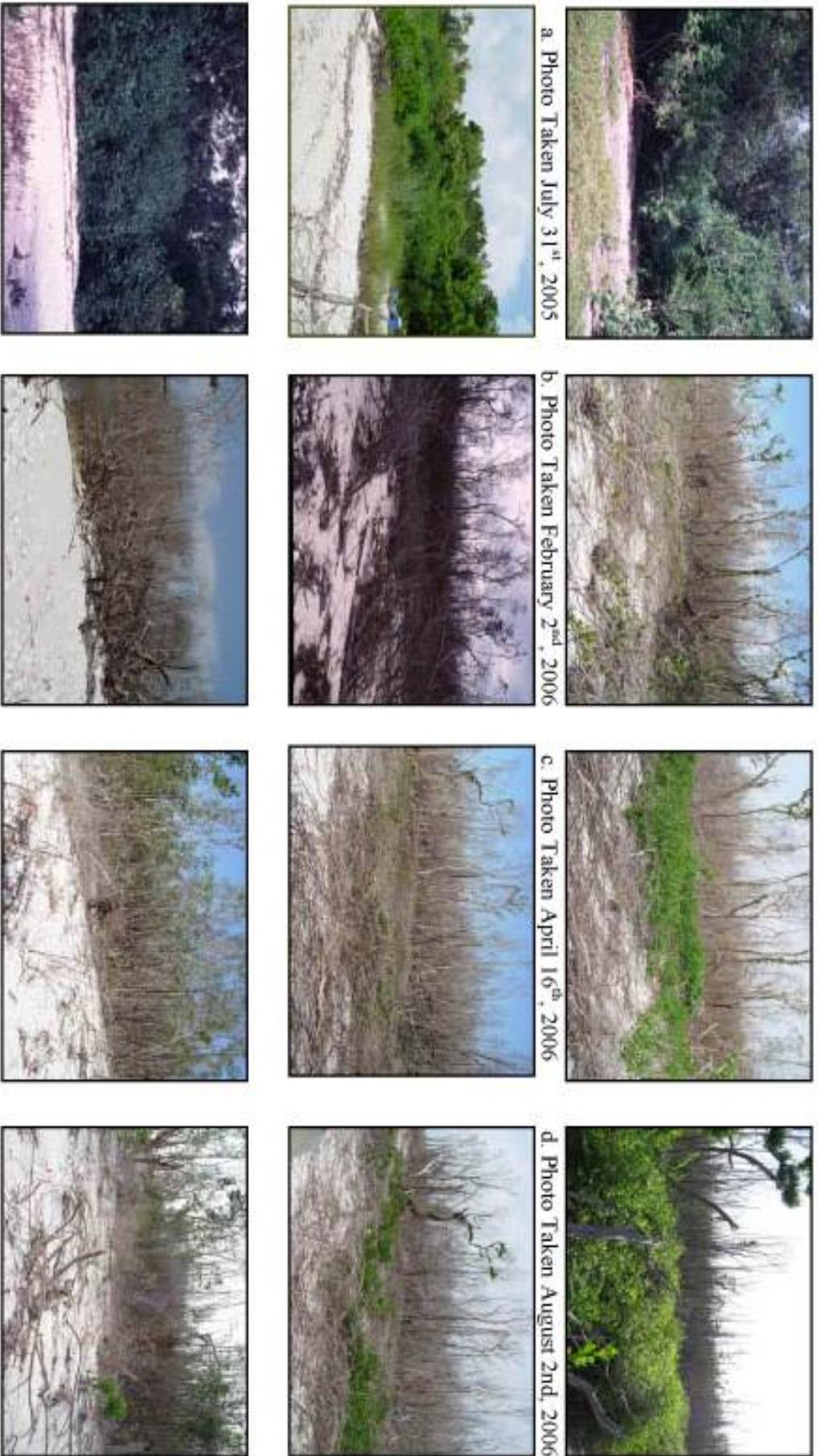


Figure 12. Vegetation communities in the lower Ten Thousand Islands were severely damaged first by Hurricane Rita and then by Hurricane Wilma. This time sequence of photographs from spatially references coordinates (+/- 2 to 5 meters) demonstrates the ability of *Colubrina* to rebound following a major storm event, relative to native species. Turkey Key (top), Wood Key (center) and Plover Key (bottom) are depicted. Photos in each column were acquired on the same date.

Allelopathy

To address speculation that chemically-mediated competition may be a primary mechanism contributing to successful invasion of *Colubrina*, McCormick and Langeland (2007) tested the effects of aqueous solutions derived from *Colubrina* leaf and root tissues on two native species. They prepared four treatments: 0.1% and 0.01% aqueous solutions of pulverized fresh leaves; 0.01% solution containing dried root tissue, and addition of freshly cut root segments to soil. The first three solutions were applied as irrigation treatments to two native species, seaoxeye (*Borricea frutescens*) and live oak (*Quercus virginiana*) seedlings. Pairs of *Colubrina* root segments were buried in cardinal directions around the stems of treatment plants, for a total of 8 root segments. Plants were watered every other day for a period of three months. Results show that growth of both species were independent of all watering treatments except for the fresh leaf solutions ($p = 0.0027$ and 0.0369 , respectively) (Table 4). Both species experienced pronounced overall loss of biomass (mostly leaf senescence and root mass atrophy or mortality) or severely stunted growth when watered with a 10% leaf solution (figure 17), but live oak also lost biomass from treatments of 0.1% fresh leaf tissue (Table 5).

Table 4. Results of greenhouse experiments testing the effects of leaf and root tissue solution and exudates on the growth of seaoxeye (*Borricea frutescens*) and live oak (*Quercus virginiana*) seedlings. Values followed by an asterisk (*) indicate significance at $\alpha = 0.05$.

SEAOXEYE (<i>BORRICHEA FRUTESCENS</i>)		LIVE OAK (<i>QUERCUS VIRGINIANA</i>)	
<i>Treatment</i>	<i>P value</i>	<i>Treatment</i>	<i>P value</i>
1% fresh leaf solution	0.1605	1% fresh leaf solution	0.7566
10% fresh leaf solution	0.0027 *	10% fresh leaf solution	0.0369 *
1% dried root solution	0.1219	1% dried root solution	0.9480
Fresh root segments	0.4243	Fresh root segments	0.6504

Table 5. Net effect on biomass of leaf and root tissue exudates. Overall, solutions created from pulverized fresh leaves had the largest negative effect.

SEAOXEYE (<i>BORRICHIA FRUTESCENS</i>)			LIVE OAK (<i>QUERCUS VIRGINIANA</i>)		
Treatment	Net Effect on Biomass	Control	Treatment	Net Effect on Biomass	Control
1% fresh leaf solution	+13.8 g	+14.7 g	1% fresh leaf solution	-4.9 g	+16.4 g
10% fresh leaf solution	-15.8 g	+3.3 g	10% fresh leaf solution	-16.9 g	+23.9 g
1% dried root solution	+15 g	+18.1 g	1% dried root solution	+10.1 g	+13.7 g
Fresh root segments	+12.9 g	+22.3 g	Fresh root segments	+19 g	+23.2 g

Though interesting, the methodologies employed in this study are simplistic and should not be extrapolated to “realistic” in-situ scenarios. Under natural circumstances, leaf litter leachate would not reach the high concentrations (0.10%) that were utilized in this study. Furthermore, leachate formation through an epigeal zone occurs rapidly and passively. In this study, secondary compounds were liberated from freshly ground pulverized leaves. Nevertheless, it is interesting to note that, even at relatively “high” concentrations of 0.01%, neither leaf nor root exudates or parts, appeared to have significant negative impacts on native species’ growth. This work suggests that chemically-mediated interference competition is not a significant mechanism responsible for the successful invasion of *Colubrina* in south Florida.

Impacts to Cultural Resources

In Everglades National Park and the Ten Thousand Islands National Wildlife Refuge, *Colubrina* invades remote key islands, some of which contain significant anthropological and archeological resources, such as Native American middens. These midden sites are often located in coastal conducive to *Colubrina* invasion, and due to their slight difference in elevation, soil temperature and soil composition, may provide ideal microsite conditions for the establishment. established, a dense thicket of *Colubrina* could compromise the structural integrity of the middens with its large root biomass, or render access to these sites virtually impossible, obscuring their location and the cultural/societal value of their contents.

Additionally, *Colubrina* infestations may diminish visitor experience in state parks and National Park Service units where tourists and day visitors access natural areas, by degrading scenic vistas, obscuring native plant species, and obstructing access.

Mapping Efforts

Attempts to document the spatial extent of *Colubrina* in south Florida at the landscape level geographic scales have been conducted in Everglades National Park. In 1981, Olmstead *et al.* conducted detailed vegetation surveys between Flamingo and Joe Bay in the Park and concluded that monotypic stands of *Colubrina* occupied less than 50 hectares (120 acres), but that these infested areas appeared to “locally engulf stands of buttonwood.” Using 1987 color infrared (CIR) aerial photographs at 1:10,000-scale, Rose (1988) analyzed the same geographic area as previously surveyed by Olmstead *et al.* and determined that *Colubrina* had expanded its range to almost 230 hectares (552 acres).

Welch *et al.* (1999) mapped vegetation communities in the Greater Everglades Region using a combination of 1:40,000-scale USGS National Aerial Photograph Program (NAPP) color infrared (CIR) 9” x 9” transparencies viewed under a stereoscope and 1:10,000-scale print enlargements. Both data sources were manually interpreted by experienced photo interpreters with expertise in Everglades vegetation communities. Extensive ground truth information collected by helicopter and airboat was employed to verify the identification of plant communities. Using a minimum mapping unit (MMU) of 1 hectare, their analysis of vegetation communities along the Florida Bay between Flamingo and Joe Bay indicate that *Colubrina* infestations increased to 302 hectares (729 acres). Decreasing the MMU to detect patches less than 1 hectare in size of either monotypic patches or *Colubrina*-hardwood vegetation mixes increased the infestation extent to 537 hectares (1289 acres). Based on mapping efforts for the entire Everglades Park, Welch *et al.* detected a total of 430 hectares (1032 acres) of monotypic (i.e., not mixed vegetation classes) *Colubrina*. Based on these data and previous mapping efforts in the Florida Bay region, Jones (1996) concludes that *Colubrina* has doubled its range of infestation every 8-10 years.

Hirano *et al.* (2003) used hyperspectral image data acquired by the Airborne Visible/Infrared Imaging Spectrometer (AVIRIS) to detect *Colubrina* in the Madeira Bay area in an effort to investigate the feasibility of using hyperspectral data to accurately detect and discriminate wetland vegetation species and communities. AVIRIS data consists of 224 discrete bands, each with 0.01-mm spectral resolution and 20-meter spatial resolution. They detected 199 hectares of *Colubrina* in patches 20 m or greater, which were primarily confined to the “buttonwood embankment”, a coastal ridge averaging 0.5 m in height separating the peninsula of Florida from Florida Bay (Figure 13). The resulting data was tested for thematic accuracy using a pre-existing Everglades vegetation database developed by Welch *et al.* (1999) and reported as 100%, indicating a 1:1 correspondence of classification agreement between these databases.

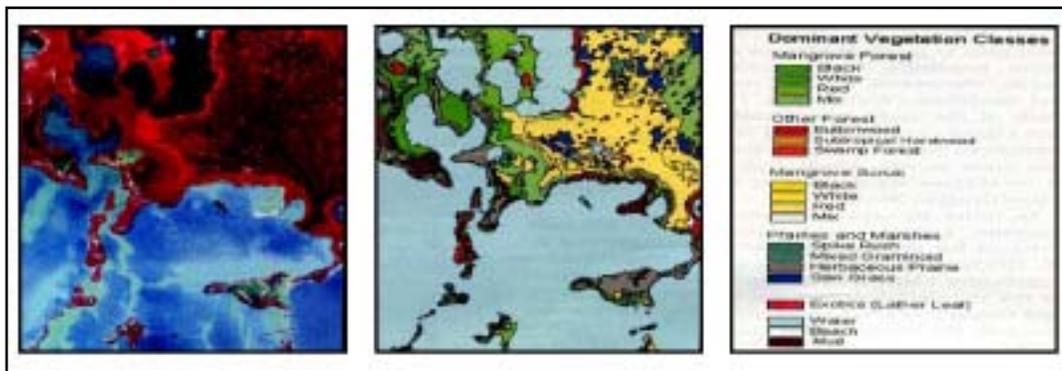


Figure 13. (left) AVIRIS image acquired over Madeira Bay and displayed as a false color image using bands 20, 30, and 42, and (center) the corresponding vegetation portion of the Everglades Vegetation Database derived from 1:40,000-scale color infrared photographs. Corresponding vegetation classification with exotics (i.e., latherleaf) indicated in magenta (right) (from Hirano *et al.*, 2003).

Management

Compiling and integrating existing data and information about *Colubrina* can assist land managers in monitoring existing populations and to identify and prevent invasions in those habitats that are vulnerable to invasion by *Colubrina* before expensive control and eradication strategies become necessary (Rejmànek and Pitcairn, 2002).

Management strategies should be firmly entrenched on local population assessments as well as community and ecosystem level impacts, rather than on “species notoriety”. Poorly planned and implemented control practices, coupled with a lack of basic information on non-native plant biology and ecology, increases the likelihood that control practices will fail or lead to non-target impacts. Such practices often have little significant, long-lasting impacts on the target species, and diminish critical resources available for future control efforts (Louda *et al.*, 1997; Pearson and Callaway, 2005). Developing a priority scheme to identify those populations that are most likely to have significant impacts is the best way to optimize the effectiveness of limited resources.

When designing control programs, land managers should consider those factors that promote ecosystem invasion, such as disturbance regime, fluctuation in resource availability and increased propagule pressure (Mack *et al.*, 2000). Ignoring such considerations will ensure that the target species continues to expand its range into new areas, and that control strategies will be subject to recurrent failure (Hobbs and Humphries, 1995). Directing resources toward understanding the mechanisms of invasion will assist managers in developing control programs that are effective at decreasing target species populations, and which are sustainable, minimize non-target impacts, and less likely to facilitate future invasions (Smith *et al.*, 2006).

Regulatory Status:

Colubrina is not considered to be problematic throughout its native range and consequently commerce and traffic in the species remains unregulated in these areas; consequently, one can readily purchase available seed via the Internet (www.b-and-t-world-seeds.com).

The species was designated a Florida Exotic Pest Plant Council (FLEPPC) “Category I” species list in the early 1990”s and was officially added to the Florida Department of Agriculture and Consumer Services Division of Plant Industry (FDACS-DPI) Noxious Weed List in March 2006. This legislation (Title XXXV of the Florida State Statute, Chapter 581, Section 091) deems it “*unlawful for any person to knowingly sell, offer for sale, or distribute any noxious weed, or any plant or plant product or regulated*

article infested or infected with any plant pest declared, by rule of the department, to be a public nuisance or a threat to the state's agricultural and horticultural interests" (Florida Senate, 2002). This state law mandates that FDACS-DPI, in conjunction with faculty from the Institute of Food and Agricultural Sciences at the University of Florida (IFAS-UF), shall review the official state lists of noxious weeds and invasive plants on a biennial basis.

In addition to state legislation, county ordinances may also prohibit the use of invasive plant species. For example, Monroe County prohibits the use of *Colubrina* as landscape material north of the Seven Mile Bridge (Municipal Code Corporation, 1984).
Miami-Dade County

Chemical Control:

The use of herbicide to control *Colubrina* is confounded by its dense, low, thicket-like growth habit and extreme difficulty in identifying the primary trunk of any given plant (Langeland, 1990). Cut-stump application of herbicide reportedly does not result in complete mortality; rather, the plant is damaged only to the point where it is re-rooted by ground layering (Schultz 1992). Langeland and Stocker (2001) recommend a basal bark application of 20 percent Garlon 4, cut stump treatment with 50 percent Garlon 3A, or foliar application with 3 percent Garlon 3A or Garlon 4 in water with surfactant. Herbicide is administered with hand-held applicators or a backpack sprayer, directly to the bark around the circumference of each stem/tree up to 40 cm (approximately 15 inches) above the ground (Langeland and Stocker, 2001). Treatments should be repeated for 3 to 4 weeks. Seedlings should be hand-pulled.

Mechanical Control:

Though unconventional, The Nature Conservancy, Florida Keys Office reports success in controlling large stands of *Colubrina* using a "weed whacker" equipped with a blade attachment (A. Higgins, *pers. communication*), rather than the standard chainsaw method. Using this technique, a technician may operate the equipment while moving through the low, sprawling *Colubrina* canopy, allowing for more effective dismemberment and removal of the stand, followed by targeted application of herbicide

to the cut stump surfaces. Re-treatment effort is cited to be substantial due to the higher probability of omitting stems during initial removal, but rapidity and reduced intensity of labor involved during initial reduction of stand biomass is reported as a significant long-term advantage in controlling *Colubrina*.

Natural Enemies

To date, no microorganism pathogens have been reported to attack *Colubrina* exclusively. Zheng *et al.* (2005) report that three arthropods are currently being tested for host specificity in China (Table 6). Of these, the long-horn beetle *Artimpaza argenteonotata* may be host-specific to *Colubrina*. Discussions are currently underway between the National Park Service (NPS) and the CABI Bioscience Centre, Switzerland to conduct a feasibility study to determine whether *Colubrina* is a suitable candidate for biocontrol (Hinz, *pers. communication*).

In south Florida, *Colubrina* may not be an ideal candidate for biocontrol efforts, primarily because there are three threatened endemic members of the *Colubrina* genus represented in the region.

Table 6. Three candidate arthropod species for host-specificity testing in China for use in classical biological control of *Colubrina asiatica* (adapted from Zheng *et al.*, 2005).

ORDER	FAMILY	SPECIES NAME
Coleoptera	Cerambycidae	<i>Artimpaza argenteonotata</i> Pic
		<i>Niphona parallela</i> White
Hemiptera	Plataspidae	<i>Paracopta duodecimpuctatum</i> (Germar)

Case Studies

Everglades National Park

Jonathan Taylor; 40001 State Road 9336, Homestead, Florida 33034; 305-242-7876
Jonathan_E_Taylor@nps.gov

Everglades National Park, a World Heritage Site and Biosphere Reserve, is comprised of approximately 700,000 acres and is the only subtropical wilderness in the continental United States. The Park was established in 1947 to conserve and protect the natural, historic, and recreational values therein. With regard to the management of invasive plant species, the National Park Service (NPS) mandates that “control, including eradication, shall be undertaken wherever such species threaten park resources, with the highest priority given to exotic species that have a substantial impact on park resource and that can reasonably be expected to be successfully controlled.” The invasive plant species of greatest concern to Park staff include Old World climbing fern (*Lygodium microphyllum*), Australian pine (*Casuarina equisetifolium*), Brazilian pepper (*Schinus terebinthifolius*), *Colubrina* (*Colubrina asiatica*), mahoe (*Hibiscus tiliaceus*), and sisal hemp (*Agave sisalana*)

Colubrina was first documented in Everglades National Park in 1954 by Frank Craighead, who noted its presence on “Crocodile Road”, most likely referring to Crocodile Point, the northern boundary of Florida Bay. By the 1970’s Park botanists had documented its spread into many other areas, noting that it was “common along the north east Florida Bay”. There were small-scale efforts to control its spread, though it was not a resource management priority. By the 1990’s, the species was documented as “common in both Florida Bay and the Ten Thousand Islands”, which prompted Park staff to seek funding for treatment programs to control *Colubrina* range expansion. Funding for treatment was not obtained, however, because at that time it was not considered a high priority. By 2002, funds were secured to initiate treatment in limited areas in the Ten Thousand Islands and Flamingo. This effort is on-going and continues to date.

In Everglades and Ten Thousand Islands, *Colubrina* occupies upper dunes, coastal strand habitat, buttonwood forests and coastal hardwood hammock. The

aggressive growth of *Colubrina* establishment and expansion in these unique and diverse habitats is of particular concern because tropical hardwood hammocks provide habitat for a number of endemic species and species of West Indian origin. *Colubrina* also establishes on storm ridges within mangrove swamps, and forms dense thickets in open/disturbed locations creating a closed canopy. The closed canopy created by *Colubrina* eliminates potential native plant recruitment. In subsequent storm events additional open sites are created which are quickly colonized by *Colubrina* resulting in more *Colubrina* acreage.

Due to the difficulty associated with detecting patches of *Colubrina* using remote sensing techniques, there have been no systematic field surveys to determine existing and potential habitat to accurately describe the cover. Fixed wing Systematic Reconnaissance Flights conducted biennially since 2002 have proven to be ineffective as a sampling technique to describe the distribution of *Colubrina*. The height and speed at which the sampling is conducted prevents accurate identification. Even lower level helicopter flights at slower speeds only picks up some of the densest populations and does not address the question of distribution.

Surveys conducted by UGA-CRMS (described in the "Mapping Effort" section) did not include the Ten Thousand Islands area. Thematic accuracy of map products were reported to be approximately 90 percent (Welch *et al.*, 1999). Additional accuracy assessments of map products by Park Service personnel have not been conducted although it is generally assumed that, based on field surveys, the estimated areal extent of *Colubrina* infestation mapped by UGA-CRM is probably under-estimated. Consequently, an accurate estimate of acreage infested by *Colubrina* in the Park remains unclear.

Manual (mechanical) removal or cutting of scandant, twining stems is employed to control *Colubrina* that impacts desirable, non-target vegetation by either over-topping it or shading it out. Young, shallow-rooted plants (seedlings and saplings) are hand-pulled intact or cut and stump-treated with a 10% solution of Garlon 4. Mature plants are treated with a basal bark or cut-stump application of 10% Garlon 4 solution. Other application methods (e.g., foliar) and herbicides (e.g., Arsenal) have been utilized with little success.

Since 2002, the annual budget for controlling exotic vegetation in the Park is approximately \$800,000. Of this amount, in-house agency funds total approximately \$35,000, with the remaining funds originating from outside sources, such as National Park Service Exotic Plant Management Team Program (NPS EPMT), Miami-Dade County, South Florida Water Management District (SFWMD), and Florida Department of Environmental Protection Bureau of Invasive Plant Management (DEP-BIPM). A summary of budgetary expenditures for *Colubrina* treatment is shown in Table 7:

Table 7. Summary of annual budgetary expenditures associated with treating *Colubrina* in Everglades National Park since 2002.

FISCAL YEAR	COST (TIME AND MATERIALS)	AREA TREATED (ACRES) GROSS INFESTED
2002	98,132	800
2003	20,000	300 re-treatment
2004	75,000	400 re-treatment
2005	28,570	300 re-treatment
2007	83,190	800 anticipated re-treat

ENP Exotic Vegetation Manager Jonathan Taylor states that, despite limited resources, successful control of *Colubrina* has been achieved in very localized areas of the Ten Thousand Islands and Flamingo. He attributes progress in these areas to diligence and an assiduous schedule of re-treatment. The inaccessibility of treatment sites and the lack of funds required to implement control regimes are cited as the primary factor that limits the success of long-term *Colubrina* treatment and eradication efforts in other portions of the Park. Additionally, Taylor cites that studies investigating the basic ecology and phenology of *Colubrina* are needed to determine whether an effective seasonal treatment regime can be applied.

Key Largo GeoPark (includes John D. Pennekamp Coral Reef State Park and Dagny Johnson Key Largo Hammock Botanical State Park – Key Largo, Florida

James G. Duquesnel, Park Biologist, John D. Pennekamp Coral Reef State Park, P.O. Box 487, Key Largo, FL 33037, 305-451-1202, james.g.duquesnel@dep.state.fl.us

Key Largo Islands GeoPark is compromised of approximately 5,475 terrestrial acres (mostly rockland hammock, coastal berm, rock barren and ruderal areas) and more than 63 square miles of submerged habitats (coral reef, mangrove swamp and other marine communities). In addition to latherleaf, park personnel also actively manage Australian pine (*C. equisetifolia*), Brazilian pepper (*S. terebinthifolius*), sapodilla (*Manilkara zapota*), beach naupaka (*Scaevola taccada*), Burma reed (*Neyraudia reynaudiana*), lead tree (*Leucaena leucocephala*), Portia (*Thespesia populnea*), laurel fig (*F. microcarpa*), lantana (*Lantana camara*), rubber vine (*Cryptostegia madagascariensis*), oyster plant (*Tradescantia spathacea*), bowstring hemp (*Sansevieria hyacinthoides*) and a few uncommon species.

The first documented observation of *Colubrina* in the park was made in 1991 by Park Biologist Jim Duquesnel, though it was certainly present prior to that time. Invaded habitats in the park include coastal berm, rock barren, rockland hammocks, ruderal shoreline and near-shore sites. Of these, coastal berm and rock barren habitats appear to be most easily invaded, as well as difficult to monitor due to the inaccessibility of these habitats. Mapping and monitoring of *Colubrina* populations is carried out by conducting on-foot transect surveys foot and recording infested areas using a GPS unit. Resulting maps are used to quantify area and density, and to prepare “project proposals”, which are sent out for bid if infestation size warrants it.

Control of *Colubrina* is generally accomplished using either Garlon 3A applied to cut-surfaces, or basal bark applications of Garlon 4. Young plants are hand-pulled, with some attention toward minimizing soil disturbance. State contractors have been assisting the Park in its *Colubrina* control efforts since 1997, and have made significant contributions toward eradication programs in the park, particularly in large infestations. Surveys are conducted by a team of technicians, capable of eradicating all but the very largest lather leaf infestations. Duquesnel (*pers. communication*) proposes variable

herbicide treatments based on stem density (number of *Colubrina* stems per unit area) in order to optimize use of limited resources available to managers (i.e., time, labor, funds) on public lands (Table 8).

Invasive plant control programs are funded almost exclusively through BIPM grants. In FY 2006, the annual budget for managing exotic vegetation was approximately \$100K, of which approximately 25% was allocated to *Colubrina* control. Park Biologist Jim Duquesnel states that progress is being made with regard to *Colubrina* control in the park, and attributes the success of this program to persistence – revisiting treated sites quarterly, and identifying previously undocumented populations and budgetary assistance from the DEP-BIPM.

Table 8. A variable herbicide treatment program based on number of stems present

# STEMS	TREATMENT	COMMENTS
< 20	Cut Stump	One applicator cuts plants close to ground level with machete (or loppers, in dense stands), followed by a second applicator who applies herbicide to exposed cambium layer. Apply herbicide immediately after cut surface for best results (figure 6).
> 20	Basal Bark	6% Garlon 4 solution in diesel fuel, as described in Langeland (1990).
> 100	Foliar	<i>Colubrina</i> over-topping other vegetation. This technique should only be administered in areas where non-target damage is minimal. Stands may require re-treatments on a quarterly basis to prevent reinfestation. Initial treatment should be confined to the perimeter of a dense stand to reduce population expansion.

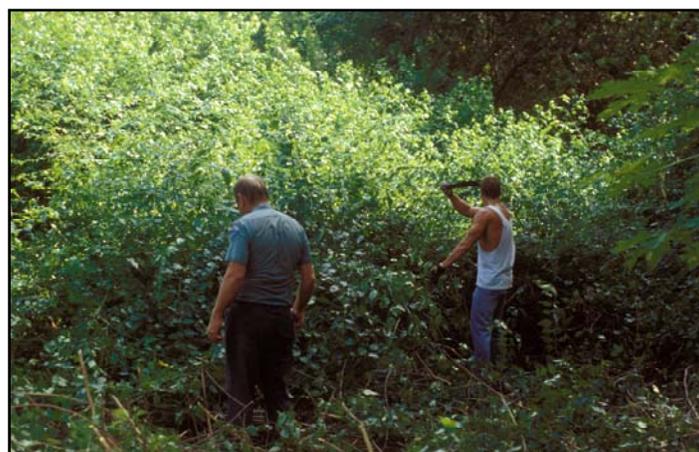


Figure 13. Figure 14. Staff from the John D. Pennekamp Coral Reef State Park removing *Colubrina* biomass close to ground level (photo courtesy of James Duquesnel).

Biscayne National Park

Tony Pernas, Florida/Caribbean Exotic Plant Management Team, 950 North Krome Avenue, Homestead FL 33030-4443, 305/224-4246, tony_pernas@nps.gov

Biscayne National Park was established in June 1980 and is located just south of Miami along the extreme southeastern coast of Florida. The 181,500 acres of the park contain a stretch of the mainland, much of Biscayne Bay, and a large number of small barrier islands off the coast. Just 30 miles from downtown Miami, Biscayne National Park contains the longest undeveloped shoreline on Florida's east coast. The park's primary vegetation communities are mangrove wetlands and hardwood hammocks on offshore islands.

Colubrina was first documented in Biscayne in the 1980's, most likely on Elliott Key and is primarily found in disturbed or open canopy areas in and adjacent to tropical hardwood hammocks on barrier islands. It is also found in disturbed areas on the mainland (e.g., jetties and storage areas). As in other Park units in south Florida, coastal hardwood hammocks are the most vulnerable to *Colubrina* invasion, as they often contain threatened and/or endangered species. The Institute for Regional Conservation (IRC) lists several species in Biscayne as being critically imperiled (below). Additionally, the state of Florida lists 65 species whose habitats include Biscayne.

- *Aristolochia pentandra* (Marsh's Dutchman's pipes)
- *Caesalpinia major* (Hawai'i pearls)
- *Guacium officinale* (Lignum vitae)
- *Opuntia corallicola* (semaphore prickly pear)
- *Phoradendron rubrum* (mahogany mistletoe)
- *Pseudophoenix sargentii* (Florida cherry palm)
- *Rhynchosia swartzii* (Swartz's snoutbean)
- *Vallesia antillana* (tearshrub)

Colubrina also out-competes nickerbean (*Caesalpinia bonduc*) which, although not a state-listed species, is an important host plant for the larval Miami blue butterfly

(*Hemiargus thomasi bethunebakeri*), which is a state-endangered species. *Colubrina* also invades sandy beaches that provide nesting habitat to several threatened and endangered species of sea turtles, most notably loggerheads (*Caretta caretta*).

Populations of *Colubrina* are mapped as they are treated. Each island is partitioned into 1-km grids; contractors sweep through each grid, treating and documenting population location with GPS coordinates. On occasion, helicopter surveys are conducted to map populations as well.

Colubrina is controlled with a cut stump application of 15% Garlon 4 in an oil adjuvant. Seedlings are hand-pulled. Biscayne National Park acquires funding for its exotic vegetation program through the National Park Service Florida/Caribbean Exotic Plant Management Team and the Florida DEP Upland Invasive Plant Program. Since FY 2000, BNP has received approximately \$100K annually.

Table 9. Annual expenditures for *Colubrina* treatment in Biscayne National Park.

Fiscal Year	Total Cost	Area Treated (acres)
2000	\$148,700	627
2001	\$130,500	614
2002	\$175,560	4381
2003	\$275,000	4936
2005	\$200,00	3698

In addition to *Colubrina*, vegetation managers also manage seaside mahoe (*Thespesia polpunea*), Australian pine (*Casuarina equisetifolia*), Hawaiian half flower (*Scaevola sericea*), Brazilian pepper (*Schinus terebinthifolius*), sapodiilla (*Manilkara zapota*), and African bowstring hemp (*Sanseveria hyacinthoides*).

Tony Pernas, coordinator for the NPS Florida/Caribbean Exotic Plant Management Team, cites aggressive treatment and re-treatment as critical elements in preventing the range expansion of *Colubrina* on BNP.

Ten Thousand Islands National Wildlife Refuge/Rookery Bay National Estuarine Research Reserve

Terry Doyle, Wildlife Biologist, Ten Thousand Island National Wildlife Refuge, 3860 Tollgate Blvd., Suite 300, Naples, FL 34114, Office: 239-353-8442 ext. 228.
terry_doyle@fws.gov

Pamela Keyes, Resource Management Specialist, Rookery Bay National Estuarine Research Reserve, 300 Tower Road, Naples, FL 34113, Office: 239-417-6310 ext. 206.
Pamela.Keyes@dep.state.fl.us

The Ten Thousand Islands National Wildlife Refuge is located approximately 20 miles southeast of Naples, Florida, on the south side of Highway 41. The Refuge is part of one of the largest expanses of mangrove estuary in North America; approximately 50% of the Refuge (18,000 acres) is mangrove forest, 9,000 acres marine water, 8,000 acres brackish marshland and other habitat (USFWS, 2006). Sections of the Ten Thousand Islands National Wildlife Refuge are co-managed with Rookery Bay National Estuarine Research Reserve. *Colubrina* was officially documented in the Refuge during a post Hurricane Andrew vegetation survey conducted jointly with the Rookery Bay National Estuarine Research Reserve (Nalley *et al.* 1997). Plant lists of the area indicate that *Colubrina* was absent from the area prior to 1992, leading biologists to speculate that viable seed was transported and deposited by Andrew along its path from the central Bahamas, across south Florida, and finally into the Gulf of Mexico. Approximately five years after its initial detection, *Colubrina* appeared to be rapidly expanding its abundance and distribution throughout the mangrove islands in the Refuge.

As in other parts of its invaded range, *Colubrina* on the Refuge and in the Reserve is found on upper high-elevation beach ridges, particularly on the outer (i.e., Gulf-side) islands. It has not been documented in the inner (i.e., mainland-side) islands. Furthermore, *Colubrina* has never been documented in tropical hardwood hammocks, except on the beach ridges, where it occasionally occurs immediately adjacent to “hammock vegetation”, where sea grape (*Cocoloba uvifera*) is very common. Biologist Terry Doyle has never observed *Colubrina* advancing inland from the beach ridge. Dense seedlings have been observed in mangrove fringes growing in unconsolidated

coralline/shell material. *Colubrina* has never been documented (either seedlings or mature plants) growing atop archaeological sites (i.e., shell mounds, middens). The greatest threat to island habitats posed by *Colubrina* in this area is the indirect effect of encroachment of and subsequent loss of, sea turtle nesting habitat and primary dune habitat.

Mature *Colubrina* plants were primarily treated using a cut-stump application of 15% Garlon 4, after which the cut wood was piled and burned. However, viable seed may become detached from branches during piling of biomass. Consequently, these seeds may require re-treatment at a later time. If stands are extremely dense, a 15% Garlon 4 solution in an oil-based adjuvant (vegetable oil) was applied in a basal bark application. Doyle suspects that additional success in eradication *Colubrina* is by incorporating fire into the treatment regime. In several instances, crews re-visit infested sites from approximately one month to nine months after initial treatment to burn the cut piles or previously treated standing dead plants with a drip torch. In addition to reducing biomass, the effect was to destroy any remaining viable seed and resprouting plants. By that time, the wood was dry and easily burned, and the leaves highly flammable. The surrounding mangrove habitats and high relative humidity from the Gulf resulted in relatively low intensity backing fire that needed no active control. Seeds remaining on the branches are incinerated, as are seeds on the soil surface subtending the stand. This method appeared to be highly effective, and Doyle reports that “not a single (*Colubrina*) seedling emerged from several large patches treated in this manner. However, outside of that patch, there may be dense patches of seedlings.” His treatment regimes identify seeds and seedlings as vulnerable life stages of *Colubrina* that should be exploited when developing control and management plans for the species.

Private contractors, funded by DEP and FWS grants, conducted initial treatments beginning in 2001, followed by DEP-BIPM funded, state-certified contractors who were responsible for follow-up maintenance (Table 3). Now that *Colubrina* has been initially controlled and in a maintenance treatment regime on Refuge and Reserve lands, in-house crews are able to monitor and treat new populations as they develop. The Refuge relies in part upon BIPM funds for their exotic vegetation programs, although no

projects ranked very high in funding priority this fiscal year. The annual budget for exotic vegetation management on the Refuge averages out to be about \$125K, of which perhaps 10% is appropriated to *Colubrina* control.

Table 10. Annual expenditures of exotic plant control funds on the Ten Thousand Islands NWR and adjacent Rookery Bay National Estuarine Research Reserve (Doyle, *pers. communication*).

Fiscal Year	Management Comment
01	On contract, had 74 acres of <i>Colubrina</i> treated for \$36K
02	Continued this work on contract for 15 acres for \$8K
02	Maintenance work on 89 acres by contract for \$14.9K
02	Also conducted in-house treatment of approx. 11 acres (no cost est.)
05	Maintenance work on 87 acres by contract for \$45K

Doyle describes the *Colubrina* program in the Ten Thousand Islands as a significant success, in part due to cooperation with agency partners such as the Rookery Bay National Estuarine Research Reserve during initial and maintenance treatments. The ability to acquire funding from both federal (USFWS) and state sources (DEP-BIPM), ensured continuity in treatment schedules, and predictability of available resources. Additionally, the ability to visit areas comprehensively, in order to estimate risk and prioritize treatment areas. Doyle targets small infestations for in-house treatment, and designates large infestations for treatment by state-certified contractors.

The key to managing *Colubrina*, according to Doyle, is to monitor infested sites regularly – “*You’ve got to keep up with it. There are no shortcuts to success*”.



Figure 14. Terry Doyle cuts and piles *Colubrina* at Round Key in March, 2000. Biomass is allowed to dry and is subsequently burned in April, 2000 (photo courtesy of Terry Doyle).

References

- Adams, C.D. 1972. *Flowering Plants of Jamaica*. University of the West Indies, Mona.
- Agren, J., 1996. Population size, pollinator limitation and seed set in the self-compatible herb *Lythrum salicaria*. *Ecology*, 77: 1779-1790.
- Armentano, T.V., D.T. Jones, M.S. Ross, and B.W. Gamble, 2002. Vegetation pattern and process in tree islands of the southern Everglades and adjacent areas.. Pp. 225-281 In: van der Valk, A. and Sklar, F. eds. *Tree Islands of the Everglades*. Kluwer Academic Publishers, Boston, Massachusetts, 541 pp.
- Association of Official Seed Analysts (AOSA), 2000. Tetrazolium Testing Handbook. Publication no. 35. Association of Official Seed Analysts, Lincoln, NE.
- Austin, D.F., 1999. Ethnobotany of Florida's Weedy Vines. *Proceedings of the 1998 joint symposium of the Florida Exotic Pest Plant Council and the Florida Native Plant Society*. Edited by David T. Jones and Brandon W. Gamble. Florida Exotic Pest Plant Council.
- Briggs, D. and S.M. Walters, 1997. *Plant Variation and Evolution*. Third Edition. Cambridge University Press, Cambridge, United Kingdom, 512 p.
- Brizicky. 1964. The Genera of Anacardiaceae in the Southeastern United States. *Journal of American Arboriculture*, 43: 359-375.
- Brown, F.B.H., 1935. *Flora of Southeastern Polynesia*, III. Dicotyledons. Bishop Museum Bulletin, 130: 1-386.
- Burkill, I.H., 1966. *A Dictionary of the Economic Products of the Malay Peninsula*. Second Edition. Kuala Lumpur, Malaysia, Ministry of Agriculture and Cooperatives.
- Carlquist, S., 1966. The biota of long-distance dispersal, III. Loss of dispersibility in the Hawaiian flora. *Brittonia*, 18:310-335.
- Carlquist, S., 1974. *Island Biology*. Columbia University Press, New York.
- Clarke, W.C., and R.R. Thaman, 1990. Pacific Island Agrosilviculture Systems: A Basis for Sustained Development and Environmental Protection. University of the South Pacific, Suva, Fiji. Available on Internet: <<http://www.unu.edu/unupress/unupbooks/80824e/80824E01.htm>>
- Coile, N., 2001. *Notes on Florida's Endangered and Threatened Plants*. Florida Department of Agriculture and Consumer Services, Contribution 38, Second Edition, Gainesville, Florida.

- Colard, J.M., F. Compennolle, and P.A. Dumont, 1976. Contribution to the study of saponins of *Colubrina arborescens* Mill: isolation of Colubrinoside and the establishment of the structure of the side chain. *Belgian Journal of Pharmacology*, 31:2-199-214.
- Crall, A.W., L.A. Meyerson, T.J. Stohlgren, C.S. Jarnevich, G.J. Newman, and J. Graham, 2006. Show me the numbers: what data currently exist for non-native species in the USA? *Frontiers in Ecology and the Environment*, 8(4): 414-418.
- Crawley, M. J. 1987. What makes a community invasible?. In A. J. Gray, M. J. Crawley, and P. J. Edwards, eds. *Colonization, Succession and Stability*. Oxford: Blackwell Scientific. Pp. 429–453.
- Culliney, John L., and Bruce P. Koebele. 1999. *A native Hawaiian garden: how to grow and care for island plants*. Honolulu: University of Hawai'i Press. p. 72-75.
- Degener, O., 1946. Tropical Plants the World Around, III.. *Journal of the New York Botanical Gardens*, 46: 132-143.
- Doren, R.F., R. Whittaker; and A.M. LaRosa. 1991. Evaluation of fire as a management tool for controlling *Schinus terebinthifolius* as secondary successional growth on abandoned agricultural land. *Environmental Management*. 15(1): 121-129. [14594]
- Duquesnel, J., 2005. Personal communication regarding treatment and control of *Colubrina asiatica* on state property in the Florida Keys. James Duquesnel, Park Biologist, John D. Pennekamp Coral Reef State Park, FL.
- FAO, 2000. Food and Agriculture Organization of the United Nations Workshop – Data Collection for the Pacific Region, FRA-WP-51. Available on Internet: <http://www.fao.org/documents/show_cdr.asp?url_file=//docrep/006/ad672e/ad672e16>.
- Florida Exotic Pest Plant Council, 1996. Category I List of Invasive Plant Species. On Internet at: < <http://www.fleppc.org/list/05List.htm>>.
- Florida Senate, 2002. Title XXXV of the Florida State Statute, Chapter 581, Section 091: *Noxious weeds and infected plants or regulated articles; sale or distribution; receipt; information to department; withholding information*. Available on Internet:<<http://www.flsenate.gov/statutes/index.cfm>>
- Ghazanfar, S.A., G. Keppel, and S. Khan, 2001. Coastal vegetation of small islands near Viti Levu and Ovalau, Fiji. *New Zealand Journal of Botany*, 39: 587-600.
- Gimlette, J.D., 1929. *Malay Poisons and Charm Cures*. Third Edition, London, England. 301 p.

- Godfrey RK, and Wooten JW. 1981. Aquatic and Wetland Plants of the Southeastern United States: Dicotyledons. Univ. Georgia Press, Athens, GA.
- Guppy, H.B., 1906. *Observations of Naturalist in the Pacific, 1896-1899*. Volume 2: Plant Dispersal. Macmillan and Company, London, England.
- Higgins, A. 2006. Personal communication via electronic mail regarding control of *Colubrina asiatica* on property own by TNC in the Keys. Alison Higgins, Land Conservation Program Manager, The Nature Conservancy of the Florida Keys.
- Hillebrand, W., 1888. *Flora of the Hawaiian Islands*. Williams and Norgate Publishers, London, England.
- Hinz, Harriet, 2006. Senior Project Scientist, Weed Control. CABI Bioscience Centre Switzerland. Email correspondence dated May 3, 2006.
- Hirano, A., M. Madden, and R. Welch, 2003. Hyperspectral image data for mapping wetland vegetation. *Wetlands*, 23(2): 436-448.
- Hobbs, R. J., and S. E. Humphries. 1995. An integrated approach to the ecology and management of plant invasions. *Conservation Biology*, 9:761-770.
- Hounsel, E.F., 2001. NMR of Carbohydrates, Lipids, and Membranes. *Nuclear Magnetic Resonance*. The Royal Society of Chemistry, 30:360-177.
- Jennersten, O., and S.G. Nelssen, 1993. Insect flower visitation frequency and seed production in relation to patch size in *Viscaria vulgaris* (Caryophyllaceae). *Oikos*, 68: 283-292.
- Johnston, M.C., 1971. Revision of *C. asiatica* (Rhamnaceae). *Brittonia*, 23(1): 2-53.
- Jones, D.T., 1996. The Status and Management of *Colubrina asiatica* (Latherleaf) in Everglades National Park. South Florida Natural Resources Center, Everglades National Park, Homestead, Florida.
- Jones, D.T., 1997. Ecological consequences of *Colubrina*(*Colubrina asiatica*) in southern Florida. *Wildland Weeds*. Winter, 1997.
- Kar, N.A., and S.R. Jain, 1971. Antibacterial evaluation of some indigenous medicinal volatile oils. *Plant Foods for Human Nutrition (Formerly Qualitas Plantarum)*, 20(3): 231-237.
- Kar, N.A., M.K. Menon, and C.S. Chauhan, 1970. Pharmacological investigations of the essential oil of *Colubrina asiatica*. *Planta Medica*, 18:222-226.

- Kingston, N., S. Waldren, and U. Bradley, 2003. The phylogeographical affinities of the Pitcairn Islands – a model for south-eastern Polynesia? *Journal of Biogeography*, 30:1311-1328.
- Langeland, K. (ed), 1990. *Exotic Woody Plant Control*. IFAS Circular 868. University of Florida Press, Gainesville, Florida.
- Langeland, K.A., and K.. Craddock Burks, 1998. *Identification & Biology of Non-Native Plants in Florida's Natural Areas*. IFAS Publications, University of Florida, Gainesville, Florida, 165 p.
- Langeland, K.A., and R.K. Stocker, 2001. *Control of Non-Native Plants in Natural Areas of Florida*. Second Edition. IFAS Publication SP-242. IFAS Publications, University of Florida, Gainesville, Florida.
- Lee, S.S., W.C. Chen, and C.H. Chen, 2000. New jububogenin glycosides from *Colubrina asiatica*. *Journal of Natural Products*, 63: 1580-1583.
- Legere, K., 2003. Plant names from North Zanzibar. *Africa & Asia*, 3: 123-146.
- Lodge, D.M., S. Williams, H. MacIsaac, K. Hayes, B. Leung, L. Loope, S. Reichard, R.N. Mack, P.B. Moyle, M. Smith, D.A. Andow, J.T. Carlton, and A. McMichael. 2006. Biological invasions: recommendations for policy and management [Position Paper for the Ecological Society of America]. *Ecological Applications* 16:2035-2054.
- Long, R.W., and O. Lakela, 1971. *A Flora of Tropical Florida*. University of Miami Press, Coral Gables, Florida, 962 p.
- Louda, S. M., D. Kendall, J. Connor, and D. Simberloff. 1997. Ecological effects of an insect introduced for the biological control of weeds. *Science* 277: 1088-1090.
- Mack, R.N., Simberloff, D. Lonsdale, H. Evans, M. Clout, and F. Bazzaz, 2000. Biotic invasions: causes, epidemiology, global consequences, and control. *Ecological Applications*, 10: 689-710.
- Marutani, M., and R. Muniappan, (1991). Succession of vegetation after suppression of *Chromolaena odorata* by *Pareuchaetes pseudoinsulata* in Guam. Proceedings from the Second International workshop on the biological control and management of *Chromolaena odorata*, Bogor, Indonesia, February 4 - 8, 1991. Available on Internet: <<http://www.ehs.cdu.edu.au/chromolanea/2/2mari.html>>.

- McCormick, C.M., and K.A. Langeland, 2007. *Seed Ecology, Allelopathy, and Post-Hurricane Recovery of Colubrina asiatica (Rhamnaceae) "Lather leaf" In Coastal South Florida*. Final Report to the National Park Service, NPS Agreement Number H5000-02-0433. Center for Aquatic and Invasive Plants, University of Florida, 28 p.
- Medan, D., 1985. Fruit morphogenesis and seed dispersal in the Colletieae (Rhamnaceae). I. The genus *Discaria*. *Botanische Jahrbücher für Systematik, Pflanzengeschichte und Pflanzengeographie*, 105: 205-262,
- Medan, D., 1991. Reproductive phenology, pollination biology, and gynoecium development in *Discaria americana* (Rhamnaceae). *New Zealand Journal of Botany*, 29: 31-42.
- Medan, D., and H.H. Hilger, 1992. Comparative flower and fruit morphogenesis in *Colubrina* (Rhamnaceae) with special reference to *C. asiatica*. *American Journal of Botany*, 79(7):809-819.
- Menges, B.S., 1991. Seed germination percentage increases with population size in a fragmented prairie species. *Conservation Biology*, 5: 158-164.
- MOBOT, 2006. Missouri Botanical Gardens *Tropicos* Database. Available on Internet at: <http://mobot.mobot.org/W3T/Search/vast.html>.
- Mogea, J.P., 2000. Understory plants and palms of the Wapoga River Area, Irian Jaya, Indonesia. Pgs. 37-38 in, *RAP Bulletin of Biological Assessment Fourteen*. Published by Conservation International, Rapid Assessment Program.
- Morton, J.F., 1981. *Atlas of Medicinal Plants of Middle America, Bahamas to Yacatan*. Springfield, Illinois. http://www.paddleasia.com/tropical_plant_edible.htm.
- Municipal Code Corporation, 2006. Code of Metropolitan Miami-Dade County, Volume I. Ordinance No. 06-115 (Supplement No. 51), enacted July 18, 2006. Chapter 18A: Miami-Dade County Landscaping, Section 18A-2H: Purpose and Intent. Available on Internet: <<http://library2.municode.com:80/mdc/home.htm>>
- Municipal Code Corporation, 2006. *Monroe County Code*. General Ordinances of the County, Ordinance No. 022-2006, (Supplement No. 91), enacted June 21, 2006. Division 10. Landscaping, Section 9.5-364. Landscaping installation criteria. Available on Internet: <<http://library2.municode.com:80/mcc/home.htm>>
- Myers, R.L., and J.J. Ewell, 1990. *Ecosystems of Florida*. University of Florida Press, Gainesville, Florida, 765 pg.

- Nalley, N., E. Langston, and T. Hopkins, 1997. Resource damage assessment of the Ten Thousand Islands National Wildlife Refuge following hurricane Andrew. Florida Department of Environmental Protection, Rookery Bay National Estuarine Research Reserve, Naples, FL, 155pp.
- National Park Service, 2006. Hole in the Donut Restoration center. On the Internet at: <<http://everglades.fiu.edu/hid/index.htm>,>.
- Nikapitiya, K., 2005. Lecture notes from *Sri Lankan Technology* (EF502), a graduate seminar offered at the University of Peradeniya, Sri Lanka. Available on Internet at: <http://www.pdn.ac.lk/eng/ocm/pdf/EF502_Lecture_1.pdf>.
- Obata, John K. 1967. Seed germination in native Hawaiian plants. *Newsletter of the Hawaiian Botanical Society* 6 (3):13-20.
- Paddle Asia, 2006. Paddle Asia, Phuket, Thailand. *Thailand tropical plants: edible plants and medicinal plants of southern Thailand*. Available on Internet at: <http://www.paddleasia.com/tropical_plant_edible.htm>
- Pearson, D. E., and R. M. Callaway. 2005. Indirect non-target effects of host-specific biological control agents: implications for biological control. *Biological Control*, 35:288-298.
- Perry, L.M., and J. Metzger, 1980. *Medicinal Plants of East and Southeast Asia: Attributed Properties and Uses*. Massachusetts Institute of Technology Press, Cambridge, Massachusetts, 490 p.
- Petelot, A., 1952. *Les Plantes Medicinales du Cambodge, du Laos et du Viet-nam*. I. Arch. Rescherches Agron. Cambodge, Laos, Vietnam, 14: 1-408; II. Arch. Recherches Agron. Pastor. Vietnam 18: 1-284. 1953; III. Ibid. 22: 1-347. 1954; IV. Ibid. 23: 1-307. [Indices].
- Pimental D., L Lach, R. Zuniga, and D. Morrison, 2000. Environmental and economic costs of nonindigenous species in the United States, *Bioscience* 50: 53-65.
- Quisumbing, E., 1951. Medicinal Plants of the Philippines. Philippines Department of Agriculture and Natural Resources, Technical Bulletin No. 16, 1234 p.
- Rejmanek, M., 1989. Invasibility of plant communities. Pages 369–388 in J. A. Drake, H. A. Mooney, F. di Castri, R. H. Groves, F. J. Kruger, M. Rejmanek, and M. Williamson, editors. *Biological invasions: a global perspective*. John Wiley and Sons, Chichester, United Kingdom.

- Rejmanek M, and M.J. Pitcairn MJ. 2002. When is eradication of exotic pest plants a realistic goal? In: Veitch CR, Clout MN (eds.). *Turning the Tide: the Eradication of Invasive Species*. IUCN SSC Invasive Species Specialist Group. IUCN, Gland, Switzerland and Cambridge, UK. p 249—53.
- Rigamoto, R.R., and A.P. Tyagi, 2002. Pollen fertility status in coastal plant species of Rotuma Island. *South Pacific Journal of Natural Science*, 20: 30-33.
- Rose, M., 1988. *Procedures: Exotic Tree Species Mapping from Aerial Photographs at Everglades National Park between Flamingo and Joe Bay*. Everglades National Park, South Florida Research Center, Homestead, Florida.
- Russell, R.P., L.L. Loope, and I. Olmstead, 1982. *Colubrina asiatica in Southern Florida*. Technical Bulletin, Report Series No. P-685. Everglades National Park, South Florida Research Center, Everglades National Park, Homestead, Florida.
- Schultz, G.E., 1992. The Nature Conservancy Element Stewardship Abstract for *Colubrina asiatica*. TNC Publication, Arlington, Virginia. Available on Internet: <URL: <http://tncweeds.ucdavis.edu/esadocs/documnts/coluasi.html>>.
- Smith, J.M.B., 1990. Drift disseminules on Fijian beaches. *New Zealand Journal of Botany*, 28: 13-20.
- Smith, M.D., J.C. Wilcox, T. Kelly and A.K. Knapp, 2004. Dominance not richness determines invisibility of tallgrass prairie. *Oikos*, 106(2): 253.
- Smith. R.G., B.D. Maxwell, F.D. Menalled, and L.J. Rew, 2006. Lessons from agriculture may improve the management of invasive plants in wildland systems. *Frontiers in Ecology and the Environment*, 8(4): 428-434.
- Spjut, R.W., 2005. Relationships between plant folklore and anti-tumor activity: an historical review. *SIDA*, 21(4): 2205-2241.
- Stafford, ,A., 2005. Personal communication regarding growth and spread of *Colubrina asiatica* following Hurricane Charlie in 2004, in an email dated August 13, 2005. Ann Stafford, Naturalist for the National Trust of Cayman Islands, Grand Cayman.
- Stohlgren, T.J., G.W. Chong, L.D. Schell, K.A. Rimar, Y. Otsuki, M. Lee, M.A. Kalkhan, and C.A. Villa, 2004. Assessing vulnerability to invasion by nonnative plant species at multiple scales. *Environmental Management*, 29(4): 566-577.
- Texas A&M University Bioinformatics Working Group, 1999. Herbarium specimen browser. On the Internet <<http://www.cSDL.tamu.edu/FLORA>>.

- Thaman, R.R., G. Keppel, D. Watling, B. Thaman, T. Gaunavinaka, A. Naikatini, B. Thaman, N. Bolaqace, E. Sekinoco, and M. Masere, 2005. Nasoata mangrove island, the PABITRA coastal study site for Viti Levu, Fiji Island. *Pacific Science*, 59: 193-204.
- Tolkachev, O.N., E.P. Nakova, R.P., 1980. Synthesis of bisbenzylisoquinoline alkaloids. *Russian Chemical Review*, 49(8): 784-798.
- Townsville Enterprise (TVE), 2006. Townsville City Council Natural Assets Database. Available on Internet: <<http://www.townsville.qld.gov.au/HabitatList.asp>>.
- Uphoff, R.A., A.M. Van den Maagdenberg, K.I. Roon, M.D. Ferrari, and R.R. Frantus, 2001. The impact of pharmacogenetics for migraine. *European Journal of Pharmacology*, 9: 1-10.
- USDA-NRCS, 2006. *Waimanalo Watershed Restoration Project: Plants to Control Stream Bank Erosion*. Available on Internet: http://www.usda.gov/stream_restoration/.
- U.S. Fish and Wildlife Service, 2006. Ten Thousand Islands National Wildlife Refuge. Available on Internet: <<http://www.fws.gov/southeast/TenThousandIsland/>>.
- Vitousek, P.M., C.M. D'antonio, L.L. Loope, M. Rejmanek and R. Westbrooks, 1997. Introduced species and global change. *New Zealand Journal of Ecology*, 12(1): 1-16.
- Wagner, H., S. Ott, K. Jurcic, J. Morton, and A. Neszmelyi, 1983. Chemistry, ¹³C-NMR study and pharmacology of two saponins from *Colubrina asiatica*. *Planta Medica*, 48: 136-141.
- Weber, E., 2003. Invasive plant species of the world: a reference guide to environmental weeds. CABI Publishing, Wallingford, UK.
- Welch, R., M. Madden, and R.F. Doren, 1999. Mapping the Everglades. *Photogrammetric Engineering and Remote Sensing*, 65: 163-170.
- Williamson M. H. 1996 *Biological Invasions*. Chapman and Hall, London, UK.
- Wunderlin, R.P., 1982. *Guide to the Vascular Plants of Central Florida*. University Press, Gainesville, Florida, 472 p.
- Wunderlin RP, and B.F. Hansen, 2002. Atlas of Florida vascular plants, Institute Systematic Botany, Univ. South Florida, Tampa, FL. Available on Internet: <URL: <http://www.plantatlas.usf.edu>>

Zheng, H., Y. Wu, Z. Ding, D. Binion, W.Fu, and R. Reardon, 2005. *Invasive Plants of Asian Origin Established in the United States and Their Natural Enemies*. Volume I. USDA Forest Service, Forest Health Technology Enterprise Team, Morgantown, West Virginia, 26505.