## Studies of the population ecology, reproductive biology and conservation status of *Crescentia portoricensis* (Britton) [Bignoniaceae]

by

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# ABSTRACT

Crescentia portoricensis Britton [Bignoniaceae] or higüero de sierra as it is commonly known (Woodbury, 1975) is an endangered shrub, strictly endemic to serpentine derived soils and to the Maricao and Susúa Forest Reserves in the western part of Puerto Rico. Due to the lack of information available on the current status of Crescentia portoricensis, an extensive search was performed for new individuals (including seedlings) and populations in likely localities in the forests and in adjacent privately owned areas. Observations on asexual reproduction in wild populations in the field were recorded. A study of the reproductive phenology was conducted. Observations on flower visitors were recorded, and plant characteristics based on Gentry's (1980) key were examined on all individuals found to assess the extent and impact of hybridization. Fruits found on the ground as well as on plants in the field were observed for dispersal, and examined for agents of fruit and seed predation. This study reports 163 and 369 plants in eight and three populations for the Maricao and Susúa Forest Reserves respectively; no seedlings were found in any of the wild populations of the species. Roots were observed to grow on stems and branches that either rested on the ground or were buried. Plants were observed to flower throughout most of the year; flowers lasted nine days on the plants. Flower visitors and evidence of hybridization were not observed. Agents of fruit and seed dispersal were not observed, but it is assumed that seeds disperse by hydrochory, and termites were also observed feeding on seeds from an old fruit that fell on the ground. This study suggests that Crescentia portoricensis is vulnerable to extinction, and its conservation status should continue to be monitored.

## RESUMEN

Crescentia portoricensis Britton [Bignoniaceae] o higüero de sierra como se le conoce comúnmente (Woodbury, 1975) es un arbusto en peligro de extinción, estrictamente endémico a suelos derivados de serpentina y a las reservas forestales de Maricao y Susúa en la parte oeste de Puerto Rico. Debido a la falta de información disponible sobre el estatus actual de Crescentia portoricensis, se realizó una búsqueda extensa de individuos nuevos (incluyendo plántulas) y poblaciones en posibles localidades en los bosques y en áreas privadas adyacentes. Se registraron observaciones de reproducción asexual en poblaciones silvestres en el campo. Se condujo un estudio sobre la fenología reproductiva. Se realizaron observaciones de visitantes florales y se examinaron las características basado en la clave de Gentry (1980) en todas las plantas encontradas, para determinar el grado y el impacto de hibridación. Los frutos encontrados en el suelo, así como los que se encontraron en las plantas en el campo, se observaron para dispersión; también fueron examinados para agentes de depredación de frutos y semillas. Este estudio reporta 163 y 369 plantas en ocho y tres poblaciones para las reservas forestales de Maricao y Susúa respectivamente; no se encontraron plántulas en ninguna de las poblaciones silvestres de la especie. Se observaron raíces creciendo en tallos y ramas que descansaban sobre el suelo o que se encontraron enterrados. Las plantas florecieron durante la mayor parte del año; las flores duraron nueve días en las plantas. No se observaron visitantes florales, ni evidencia de hibridación en las plantas. No se observaron agentes de dispersión o depredación de frutos, pero se asumió que las semillas se dispersan por hidrocoria; además, se observaron termitas alimentándose de semillas de un fruto viejo que había caído al suelo. Este estudio sugiere que Crescentia portoricensis es vulnerable a extinción, y que su estado de conservación se debe continuar monitoreando.

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# **DEDICATION**

Nunca tuve la oportunidad de despedirme y no lo voy a hacer ahora, te fuiste tan rápido y sin aviso, yo esperaba volver a verte. Yo se que la pesca es muy buena allí, que el sol está caliente y que el mar está planchao y algunas veces alborotao. Tu hija está grande, creciendo rápido y está siguiendo tus pasos, pero yo sé que tu siempre estás ahí pa' ella. Yo estoy por terminar esto y espero verte pronto, pero por ahora hay que seguir pa' lante por las nenas. Bueno, esto es pa' ti', no es lo mejor del mundo pero va por ahí. Te envío una foto, nos vemos, te veo pronto.



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# **INTRODUCTION**

*Crescentia portoricensis* Britton [Bignoniaceae] or higüero de sierra (Fig. 1) as it is commonly known (Woodbury, 1975) is an endangered shrub, endemic to the Maricao and Susúa Forest Reserves in the western part of Puerto Rico. Gentry (1980) described *Crescentia portoricensis* as a vinelike shrub with long slender branches. Plants can reach up to 6 m in height and produce hermaphroditic, yellowish-green bell-shaped flowers (Figs. 2 and 3) that ripen into dark green fruits (Fig. 4) (Little et al., 1974). It grows as an open, sparsely-branched shrub with one to five lax, wand-like basal branches. The leathery leaves occur in clusters on highly reduced short shoots along the main branches (Breckon and Kolterman, 1993). The leaves, mostly appearing in fascicles of two or three (Fig. 5), are alternate but with congested internodes. The inflorescence is a single axillary flower, with a greenish or yellowish corolla. The plants are cauliflorous, i.e., the flowers and fruits are borne along the main stems, not on smaller branches (Breckon and Kolterman, 1993).

There is a lack of information available on the ecology and reproduction of *Crescentia portoricensis*. Breckon and Kolterman, throughout their four-year study of the species (1991-1995), located reported and unreported populations, tagged the great majority of the plants, studied reproductive phenology and described the species' habitat requirements. Relatively few herbarium collections have been documented for the species. The MAPR (see Appendix I for abbreviations) herbarium's BRAMHS database includes 24 collections, of which 16 have been made since 1986. Table 1 shows the historical collections for the species, defined as those collected more than 35 years ago.

*Crescentia portoricensis* is found in the subtropical moist, subtropical wet and lower montane wet forest zones in Puerto Rico, which occupy an area of 2,124.8, 5,326.1 and 109.1

km<sup>2</sup> respectively (Ewel and Whitmore, 1973). It presently ranges from 135 to 850 m in elevation, not just between 300 and 800 m as reported by Gentry in 1980 (Breckon and Kolterman, 1994). It is a shade-tolerant and moisture-limited understory shrub, restricted to the bottoms of shady, mesic quebradas at lower elevations and extending onto more exposed slopes at higher elevations (Breckon and Kolterman, 1994). The plants are found along the banks of streams, many within only a meter of the water's edge, and are apparently restricted to the Susúa and Maricao Commonwealth Forests (Fig. 6). They occur only on serpentine-derived soils; therefore the species is a local endemic (Breckon and Kolterman, 1994). Most likely *C. portoricensis* is the most widespread of the 12 endemic serpentine-restricted plants in Puerto Rico (which consist of about 5% of the total endemics on the island), but it is never common (Cedeño-Maldonado and Breckon, 1996).

Serpentine soils are characterized to be high in the concentration of Mg in comparison with Ca, high in concentrations of heavy metals such as Ni, Cr and Co, and poor in fundamental nutrient elements such as nitrogen, phosphorus and potassium (Karataglis et al., 1982). As a consequence of these characteristics, which make the soil unsuitable for the normal growth of many plant species, the vegetation on these soils is mostly scattered, dwarfed and xerophytic (Karataglis et al., 1982).

The Maricao Forest Reserve (Fig. 7) is located at the western extreme of the Cordillera Central (Rivera et al., 1983) and was established as a state forest in 1919 (Rivera et al., 1983; Silander et al., 1986). It consists of 4,246 ha (A. Muñiz-Suárez, pers. comm.; see list of affiliations in Appendix II); 85% of its soils are serpentinitic (Silander et al., 1986). Elevation in the forest reserve ranges from 150 to 900 m (U.S.G.S. topographic map, 7.5' series, Maricao quadrangle, 1960); the average temperature is 22°C, and the mean annual precipitation is 2,540

mm (A. Muñiz-Suárez, pers. comm.). In 1973, Ewel and Whitmore reported three life zones for the forest reserve: subtropical moist forest, subtropical wet forest (the largest in extent) and lower montane wet forest. According to Little and Wadsworth (1964) and Little et al. (1974), 845 species of vascular plants can be found in the Maricao Forest Reserve, of which 278 are woody plants. Of these 278 species, 123 are endemic to Puerto Rico and 20 to Maricao (Little and Wadsworth, 1964; DNR, 1976). In the Maricao Forest Reserve five vegetation types can be found in the three bioclimatic life zones. One of these is the dwarfed vegetation of evergreen, small-leaved species that occupy the narrow ridges, peaks and summits exposed to strong winds, which are found nowhere else in Puerto Rico (DNR, 1976). The other is an exclusive element of the forest floor formed by large cushions of the rare "reindeer moss," a lichen of the genus Usnea (I. Sastre De Jesús, pers. comm.), that occupy some ridges and windward slopes (DNR, 1976). Three factors may help explain the rich diversity of trees resulting in the distinctive forest types occurring in Puerto Rico only in the Maricao Forest Reserve: closed nutrient cycling, adequate precipitation received on the well-aerated serpentine soils and the atypical combination of physiographic characteristics (DNR, 1976).

The Susúa Forest Reserve (Fig. 8) was established as a reserve in 1935 (Silander et al., 1986). In the 19<sup>th</sup> century this forest was cut due to the demand for agriculture, wood for fuel and construction (Álvarez, 1983). The forest reserve consists of 1,313.1 ha (W. Cordero, pers. comm.), and 90% of its soils are serpentinitic (Silander et al., 1986). Its elevation ranges from 80 to 473 m (Silander et al., 1986). The Susúa Forest Reserve receives a mean annual precipitation of 1,413 mm and has a mean annual temperature of 23.9°C (Álvarez, 1983). According to Ewel and Whitmore (1973), the forest reserve is located in the subtropical moist forest life zone. In 1991, García reported 296 species of vascular plants for the forest reserve, 38 of which are

endemic to Puerto Rico, with one, *Calliandra locoënsis* García & Kolterman (Leguminosae: Ingeae) restricted to the forest. According to Breckon and Kolterman (1992) the vegetation in the Susúa Forest Reserve is characterized as azonal; its poorly developed soils make it more xeric than it should be given the climate.

Within the Susúa Forest Reserve, three general habitats occur: ravines (quebradas), slopes and mountain summits (García, 1991). Along the bottoms, the ravines are mesic and shady. The slopes vary from mesic to xeric, depending on their angle, exposure and elevation. The summits are xeric, like the steep upper south- and east-facing slopes in the forest reserve (Breckon and Kolterman, 1992). González-Rodríguez (1998) states that two vegetation types have been described for this forest reserve: the dry slope forest and the gallery forest. In the dry slope forest low, xeric, shrubby species predominate; thorny species are common and cacti and other succulent species are uncommon but can be found in the herb-subshrub layer (Breckon and Kolterman, 1992). García (1991) characterizes the gallery forest as mesophytic vegetation, in which the trees can reach up to 15 m in height. This type of vegetation is found at the margins of rivers and creeks. According to Álvarez (1983), these gallery forests develop on alluvial soils classified as Quebrada silty clay, which border the margins of rivers and quebradas. Of a total of 1,313.1 ha of forest reserve land, 95.2 ha have been estimated to consist of gallery forest (García, 1991).

Breckon and Kolterman studied *Crescentia portoricensis* from 1991 to 1995. During this four-year study they located, labeled and measured individuals. According to Breckon and Kolterman (1994) 100 individuals were found in the Maricao Forest Reserve, 84 of which were observed in the forest understory on slopes along the Río Maricao. The Río Maricao population is particularly important, given the fact that the locality for the type specimen of the species

(Britton, Stevens and Hess # 2455) is cited as "Río Maricao" (Table 1). *Crescentia portoricensis* is found in the forest understory of ridgetop forests, canyons and upper slopes of ravines in the Maricao Forest Reserve. The individuals found in this forest reserve were seen to be more robust, had larger leaves and had a greater light exposure compared to the individuals in the Susúa Forest Reserve. Breckon and Kolterman indicated that the greater size of its leaves and denser foliage in association with greater light exposure may explain their robustness.

Two hundred thirty-one individuals were found in the Susúa Forest Reserve; the elevation for these individuals ranged from 135 to 250 m. Of these 231 individuals, 16 were found just outside the reserve's boundary. Crescentia portoricensis occurs in three separate canyons within the Susúa Forest Reserve; the rivers and streams that drain from these canyons become part of the Río Loco drainage (Breckon and Kolterman, 1992). Crescentia portoricensis was found to be limited to the lowest portion of the quebradas, along a shelf immediately above the high water mark. The Arroyo del Tanque group is the exception; it is located on a slope that faces northwest along a stream. In their study the number of basal branches ranged from one to five, but the majority of plants had a single basal branch. Basal stem diameter ranged from 0.5 to 9.0 cm. Vegetative reproduction was observed at the terminal portion of the lax branches coming in contact with the ground and layering; asexual reproduction showed its highest incidence in the Quebrada Grande population. Fifty-two fruits (at least 4 cm in diameter) were observed on 27 individuals. Fruits were green in color when immature; as they ripened they would turn a dull blackish gray or brown. Breckon and Kolterman (1992) found that old fruits were sometimes chewed open; it was presumed that the introduced rat could have done the damage. In some cases they found that the whole contents were eaten; and inferred that this could explain the total lack of seedlings in the wild.

In a germination experiment conducted in the University of Puerto Rico Biology Department's greenhouse in 1993, different combinations of soil moisture (wet, mesic and dry) and light intensity (shade, semishade and sun) regimes were evaluated. The highest germination rates were found under the conditions that prevail in the species' native habitat (wet to mesic soil under low to moderate light intensities). From the germination experiments and observations in the field, Breckon and Kolterman concluded that *Crescentia portoricensis* is shade-tolerant and moisture-limited. They did not conduct tests to determine the relationship between fruit condition and seed viability, although they discovered that seeds from full-size green fruits germinate readily in the greenhouse.

In 1994, Breckon and Kolterman reported that *Crescentia portoricensis* probably flowers throughout most of the year, based on a general phenological pattern of sporadic flowering. Flower buds (Figs. 9 & 10), flowers and fruits were observed on 32 individuals in the Maricao Forest Reserve. In 1993 a flower corolla was found on the ground; apparently a robber had cut a slit at its base to obtain nectar. On April 3, 1993, seven plants in Río Maricao showed asexual reproduction, plants had rooted along prostrate branches. Reproductive efforts (flowers, fruits and asexual) were only observed on the larger plants. Eight plants bore a total of eighteen reproductive structures: seven flower buds, seven flowers, and four fruits. The largest fruit they found measured 13.6 cm long x 3.2 cm in diameter.

Prior to their designation as public forests, much of the Susúa forest and parts of the Maricao forest were cut for cultivation, grazing, charcoal production and wood (USFWS, 1991). In 1493, on Columbus's second voyage, Puerto Rico was first visited by Europeans; within 40 years of their first visit, Puerto Rico had 10 sugar mills and a placer gold mine on the southern flanks of the Luquillo Mountains. Between 1830 and 1950 much of Puerto Rico was cleared for

agriculture (Clark and Wilcock, 2000). Forest cover has increased since 1950, but many of these forests are dominated by non-native species (Lugo, 2004).

*Crescentia portoricensis* is endemic to serpentine soils and is found only in two forest reserves in southwestern Puerto Rico. Its rarity and isolation are reflected in the fact that it was first described less than a century ago, and only 331 individuals in 11 populations were reported up to 1996. *Crescentia portoricensis* was listed as endangered December 4, 1987 (USFWS, 1991). According to the U. S. Fish and Wildlife Service (1991) the species will be considered for downlisting when its habitat is protected and four new populations are established. *Crescentia portoricensis* is endangered as a result of intensive deforestation and land management practices that have negatively affected its populations through flooding and erosion, which are believed to be responsible for the eradication of two previously reported populations in the Maricao forest (USFWS, 1991). Because of the increasing erosion of stream banks, biologists of the Department of Natural and Environmental Resources working in these forests have observed the loss of individuals (USFWS, 1991).

A management plan that considers the presence and requirements of this and other rare plant species has not been established for either of the two forest reserves in which it occurs. Reproductive biology, pollination and seed dispersal mechanisms need to be defined for conservation management (USFWS, 1991). Given the species' rarity, its restriction to serpentine soils and its importance as an endangered and endemic plant species, the purposes of this study were to determine the present status of the species and to obtain current information about its population ecology and reproductive biology.

7

# **OBJECTIVES**

Research will be based on seven main objectives:

- 1. Assess the present status of known populations and individuals of *Crescentia portoricensis* in the Maricao and Susúa Forest Reserves, Puerto Rico.
- 2. Search for new individuals and populations in the forests and adjacent privately owned areas.
- Gather observations on recruitment by asexual and/or sexual reproduction in wild populations.
- 4. Study the reproductive phenology (periodicity, frequency, timing, and duration of flowering and fruiting, and abundance of flowers and fruits) and gather observations on pollination.
- 5. Assess the extent and impact of hybridization.
- 6. Gather observations on seed production, fruit and seed dispersal (including possible secondary dispersal), and fruit and seed predation.
- Evaluate the conservation status of the species and make recommendations regarding its eventual recovery.

# LITERATURE REVIEW

#### Family

The family Bignoniaceae is distributed throughout the tropics; few species can be found in the temperate zone. Bignoniaceae are mostly Neotropical, in this region 620 or 78% of a world total of 800 species are native (Gentry, 1980). Brazil is the center of diversity for the family, but if the number of species is taken as a diversity criterion, then each tribe would have a different and unique center of diversity (Gentry, 1980). Gentry assigned the Neotropical Bignoniaceae to six tribes: Bignonieae, Tecomeae, Crescentieae, Schlegelieae, Tourrettieae and Eccremocarpeae. The genus *Crescentia* L. is in the tribe Crescentieae.

#### Genus

*Crescentia* is a small genus native to tropical America. It has been monographed by Gentry (1980), who describes the plants as small to medium-sized trees, with rachitic branching and an open crown. He states that the flowers are bat-pollinated and the fruit is large, more or less spherical or terete, indehiscent, with a hard woody shell and pulpy inside. There are six species in tropical America ranging from Mexico and the West Indies to Amazonian Brazil and Perú (Gentry, 1980). Characters such as leaf size, shape, and texture, corolla lobe shape, and fruit size and shape are used to define the species. Even the most distantly related species (e.g., *C. cujete* and *C. alata* Kunth) are interfertile (Gentry, 1980). Apparent hybrids between these two species have been collected in Mexico, Guatemala, El Salvador, and Costa Rica (Gentry, 1980). The species tend to form dense aggregations of homogeneous morphotypes where they occur, and taxonomic recognition of different-appearing ecologically differentiated dominants seems in

order despite the tenuous nature of their reproductive barriers and distinguishing characters (Gentry, 1980).

*Crescentia cujete*, the most commonly known of the species in the genus, is cultivated throughout most of the tropics. It propagates easily from seeds or cuttings but grows slowly (Little and Wadsworth, 1964). Since pre-Columbian times the fruits have been used to make bowls, cups, jugs, water containers, ornaments and musical instruments (Gentry, 1980; Little and Wadsworth, 1964). According to Domínguez-Cristóbal (2000), Fray Iñigo Abbad y Lasierra documented in his report (Historia geográfica, civíl y política de la isla de San Juan Bautista de Puerto Rico, published in Madrid in 1788) to the Conde of Floridablanca that the Taínos living in Puerto Rico planted *Crescentia cujete* very close to their homes because of the species' utility to them, and that the fruits from *C. cujete* were used in the elaboration of domestic artifacts such as spoons, cups and plates and the musical instrument called maracas. *Crescentia cujete* is probably native to Mexico and northern Central America, and can be found throughout Puerto Rico today (Gentry, 1980; Liogier, 1995).

*Crescentia alata* is native to Mexico and Central America, where it is normally found as a dominant plant in dry savannas. Today it can be found in Puerto Rico as an introduced species (Gentry, 1980; Liogier, 1995). It is cultivated in Cuba and the Old World tropics, where its fruits are used to make cups, rattles and ladles, but farmers prefer the fruits of *C. cujete* because of their greater size (Gentry, 1980). *Crescentia amazonica* can be found in Colombia, Venezuela, Brazil and Perú, where it inhabits the upper and central Amazon and Orinoco and their main tributaries in the seasonally flooded varzea and tahuampa forests (Gentry, 1980). Gentry (1980) believed that *C. amazonica* might be a "wild form of widely cultivated *C. cujete*." He stated that the fruits from *C. amazonica* are smaller than those of *C. cujete* and that natural selection for the small fruits might have occurred if human selection for large fruits were relaxed. He hypothesized that biogeographically the species is a bit doubtful, because members of the tribe Crescentieae primarily inhabit Central America and the West Indies. Early European explorers mention the species, so Gentry (1980) hypothesized that if the introduction of the species was not natural, it might have been pre-Columbian.

Three of the six species are essentially endemic to the Greater Antilles. Crescentia mirabilis Ekman ex Urban occurs only in northeastern Cuba (Gentry, 1980), in the coastal marshes of the Puerto Padre region of the extreme northern Oriente Province and neighboring Camagüey. Crescentia linearifolia Miers occurs in Hispaniola, Puerto Rico, the U.S. Virgin Islands and on St. Barthélemy, and is perhaps also native to Belize (Gentry, 1980). In 1995, Liogier documented that Crescentia linearifolia occurs on hillsides and in woodlands along the southern coast of Puerto Rico, from Boquerón east to Salinas, at the Cabezas de San Juan, Fajardo and on Piñeros Island (located east of Punta Medio Mundo). Piñeros Island was formerly under the jurisdiction of the U.S. Navy as part of the Roosevelt Roads military complex in Ceiba, and consists of 327.94 cuerdas (M. Justiniano, pers. comm.) or 128.9 ha. In 1993, Breckon and Kolterman reported hybrids along P.R. Hwy. 116 in the Lajas Valley, where a natural population of C. linearifolia is apparently hybridizing with the introduced C. cujete. It was reported that the majority of the trees are apparently hybrids; trees that appeared to be "pure" C. linearifolia were infrequent in the population, and the species could be in danger of being hybridized out of existence there. The other species native to Puerto Rico is Crescentia portoricensis, which Gentry believed might better be transferred to Amphitecna Miers, depending on whether the placentation is completely parietal vs. axile to axile basally and parietal above (G. J. Breckon,

pers. comm.). *Crescentia portoricensis* is endemic to southwestern Puerto Rico, restricted to serpentine soils, and considered to be both rare and endangered.

#### **Rare species**

Woodbury (1975) and Rabinowitz (1981) refer to species as being rare when they occur in small numbers, have a limited range or are restricted to a specific or specialized habitat, and an increase of pressure on them or their habitat could result in endangerment. According to Rabinowitz (1981) and Kruckeberg and Rabinowitz (1985), rare species occupy only a small portion of a floristic area or geographic province. Schemske et al. (1994) state that factors such as allele number, frequency, heterozygosity, reproductive system, birth rate, growth rate, death rate, colonization of unoccupied habitats and extinction, operating at a range of spatial scales, could be responsible for the abundance or rarity of plant species. Rarities can occupy extensive, stable, climax forest situations; their present restriction could be explained by the past history of the area in which they are found today or the destruction of their specialized habitat within the climax forest (Kruckeberg and Rabinowitz, 1985).

#### **Endangered species**

According to Woodbury (1975), endangered refers to species that usually occur in small numbers and are restricted to a specific habitat or a limited range; they are in immediate risk of extinction and without any special protective measures would not survive. The USFWS defines an endangered species as a "species which is in danger of extinction throughout all or a significant portion of its range" and is endangered as a result of the following factors: (A) the present or threatened destruction, modification, or curtailment of its habitat or range; (B) overutilization for commercial, recreational, scientific, or educational purposes; (C) disease or predation; (D) the inadequacy of existing regulatory mechanisms; or (E) other natural or manmade factors affecting its continued existence (epw.senate.gov/esa73.pdf). In 1987, Raven projected that in the next 50 years, 25% of the estimated 250,000 species of vascular plants in the world may become extinct; by 1992, 22% of the vascular plant species in the United States were of conservation concern (Falk, 1992). According to the USFWS (1988) since the passage of the Endangered Species Act in 1973, plant conservation efforts have started to improve. The Act established a legal mandate of unprecedented proportions to promote the collection, analysis, and exchange of biological information and required that, for each endangered or threatened species in the United States, a recovery plan must be developed that "delineates, justifies, and schedules the research and management actions necessary to support the recovery of a species" (USFWS, 1988).

Schemske et al. (1994) analyzed recovery plans for 91 species; the majority of the species showed a very restricted geographic range and narrow habitat requirements. States (or territories, e.g., Puerto Rico) per species averaged  $1.68 \pm 1.54$  (mean  $\pm 1$  sd; range 1-12), with 66.3% (65 species) in only one state, and 23.5% (23 species) in two states. Seventy-three species or 85.9% were found to have narrow ecological requirements, but data were only available for 85 of the 91 species. The principal causes of endangerment for the 91 species examined are: development (20.4%), over-collecting (10.2%), grazing (10.2%), oil, gas and mining (8.2%), trampling (not specified if by humans or animals) (8.2%), water control (8.2%), logging (7.1%), off-road vehicles (6.1%), exotic plants (6.1%), agriculture (5.1%), roads (4.1%), fire control (4.1%), military (1.0%) and natural impacts (1.0%). A problem with the Endangered Species Act of 1973 is that federally listed fish and wildlife (animals) are protected on all lands, public or private, but endangered plants are only protected in areas under federal jurisdiction (M. Justiniano, pers. comm.). According to Schemske et al. (1994) the authority of recovery plans becomes limited, because in their survey of recovery plans, private lands were important in the conservation of at least 50% of the federally listed species. Furthermore, in 1992, the USFWS stated that nearly 50% of the 749 federally threatened or endangered species are plants, but in 1990 endangered plant species received only 8% of recovery funds spent by the agency (Campbell, 1991). In Puerto Rico, 49 of the 79 federally threatened or endangered species are plants (http://www.fws.gov/), about 63% of the listed species, but historically they receive less attention than animals due to the fact that Puerto Rico has an emblematic animal (Puerto Rican parrot) and not an emblematic plant (M. Rivera, pers. comm.).

#### **Causes of rarity and endangerment**

Rabinowitz (1981) and Fiedler and Ahouse (1992) state that rarity and endangerment of a plant species may be caused by intrinsic factors (related to the biology of the species) or extrinsic factors (related to the environment). Although some extrinsic factors are natural, humans are the cause of a great number of them (Liu and Koptur, 2003). *Crescentia portoricensis* is endangered today, in part, as a result of extensive deforestation and poor management practices that have promoted flash-flooding and erosion, which are believed to have been responsible for the eradication of two populations in the Maricao Forest Reserve (USFWS, 1991).

In 2000, Liogier and Martorell stated that the Puerto Rican native vascular flora consists of approximately 3,000 species. Forty-nine out of 3,000 or 1.6% are listed as endangered by the

U.S. Fish and Wildlife Service and almost all of them can be found either in the northern Karst region or in the dry south-southwest portion of the island (http://www.fws.gov/). Breckon (unpublished data) estimates that approximately 30% to 40% of the species that compose the Puerto Rican flora are not native to the island (Román Guzmán, 2006). Breckon hypothesized that these species can be found on the island as a result of human activities, and that if this estimate is correct, then the percentage of native plant species that are endangered would rise to 2.3-2.7%. Unpublished data by Breckon and Kolterman show that a number of plant species should be listed as endangered and more than 60 species that were previously reported for Puerto Rico have a high chance of extinction on the island (Román Guzmán, 2006).

### **Deforestation and its consequences**

According to Tolera et al. (2008) when forests are cleared habitat loss occurs, the viability of populations is affected (diversity at the genetic level is lost), individuals of the same species become farther apart from each other (isolation of individuals) and populations become fragmented. Deforestation causes the loss of biodiversity, and changes regional hydrology, climate and terrestrial carbon storage. It also degrades the living standards of human communities, interrupts environmental services (causes loss of forests for flood control, promotes loss of soils and contributes to the development of wildfires), and affects ecotourism industries (Laurance et al., 2001).

A major consequence of deforestation is erosion. Direct effects of erosion include fragmentation of mountains, redistribution of minerals and nutrients over the surface of the land and topographic changes (Lugo et al., 1980). Another negative consequence of erosion is modification of the pathways of water and sediment transport (Conacher, 2002).

In vegetated areas the structure of the vegetation slows down erosion and the transport of sediments by the interception of rain by the forest canopy, thus reducing the strength and speed of raindrops (Bochet et al., 1998; Lugo et al. 1980). Vegetation acts as a physical barrier by modifying the flow of sediment at the surface of the soil (Lee et al., 2000; Van Dijk et al., 1996). When roots penetrate the ground they slow down erosion by increasing the structural stability of the soil (Bochet et al., 1998). Roots improve water movement, gaseous diffusion, and push aside soil particles, thus increasing bulk density of the soil near the roots (Bochet et al., 1998). Forest cover is beneficial against erosion and runoff because it produces roots, leaf litter, and enriches the ground with biota thus increasing the infiltration of rain (Gyssels et al., 2005; Körner, 2002; Puigdefábregas, 2005; Rey, 2003).

#### **Deforestation in Puerto Rico**

Before the Europeans arrived in 1493, Puerto Rico was almost 100% forested (Rudel et al., 2000; Wadsworth, 1950); Little et al. (1974) estimate that its native flora consisted of approximately 547 tree species. By the 19<sup>th</sup> century, deforestation characterized most of the island; nearly all of the trees with economic potential were cut to be used for wood and charcoal and to make space for agriculture (Lugo et al., 1980; Lugo, 2004). High levels of erosion, soil compaction and choking of river channels with sediment were caused by agriculture in Puerto Rico (Clark and Wilcock, 2000). By the 1940's only 6% of forest remained, 1% of original forest remained untouched, and the remaining land was used for agriculture (Lugo, 2004). By the 1990's, the population on the island had increased to approximately 450 people per km<sup>2</sup> and urban land cover had increased to about 14% (López et al., 2001). Along with an increase in the population came the growth in urbanization, and the disturbance changed in character; it went

from cutting and burning for agriculture and fuel to bulldozing and clearing for roads, transmission lines, ports, quarries, power plants, urbanizations, shopping malls and industrial centers (Román Guzmán, 2006). For example, urban and land development increased from 1.7% in 1951 to 15.4% in 2000 (Kennaway and Helmer, 2007).

The most dramatic event of forest recovery in the history of the world occurred in Puerto Rico; from 1950 to 2000 much of the land that had been cultivated was abandoned; the cultivation of lands declined by 95% (Kennaway and Helmer, 2007; Rudel et al., 2000). For example in 2007, Kennaway and Helmer found in their study that from 1951 to 1991 coffee with mixed and woody agriculture declined by 17%. Forest recovered as a result of a shift in the economy from agriculture to industry (López et al., 2001), and the migration of the population from rural to urban areas to work in industry. Operation Bootstrap was implemented in the 1940's in which companies that set up industrial facilities in Puerto Rico (1,400 of them established in the first two decades) were given exemptions from federal and state taxes (Rudel et al., 2000). Another reason for the increase in forest cover was the emigration to the United States (Kennaway and Helmer, 2007). Forest expansion continues today; thanks to the creation of forest reserves and reforestation efforts on the island, 44.8% of the island is presently covered by forest/woodland/shrubland of which approximately 5.2% is under protection (Kennaway and Helmer, 2007). Recently the recovery in forest cover has slowed down; it has been hindered by forest clearing for land development for urban or suburban residential, industrial, commercial, or transportation purposes (Kennaway and Helmer, 2007).

#### Asexual vs. sexual reproduction

Asexual reproduction or vegetative reproduction as it is commonly known, results in progeny that are an exact genetic replica of their single parent (Raven et al., 2005). It is frequent in plants and is produced in different ways, such as stolons or seeds produced asexually (agamospermy) (Raven et al., 2005). The populations cannot adapt to the changing conditions as well as those that can reproduce sexually, because they do not have the recombination and genetic variability necessary to do so (Liu and Koptur, 2003; Raven et al., 2005). Wild populations that are capable of invading new environments in competition with others will have the advantage (Raven et al., 2005). Asexual reproduction is important because a pollinator is not needed for reproductive structures such as buds, flowers and fruits. Also, plants can produce progeny in a short period of time from a small quantity of vegetative material (Johnson and Sindel, 2005).

#### **Reproductive phenology**

Rathcke and Lacey (1985) define plant phenology as "the study of the seasonal timing of life cycle events." Plant phenology is the complex result of several plant processes that are normally described independently, such as leaf elongation, bud formation, flowering, fruiting and seed germination (Bertiller et al., 1990; Sakai et al., 1999). Individual fitness, as well as population or species survival, can be greatly affected by the distribution over time of life-history events, such as growth, leaf flux and fall, flowering, fruiting, and seed dispersal. Studies in plant phenology seek cyclic patterns related to environmental seasonality (Madeira and Fernandes, 1999). These cyclic patterns or phenological schedules could be affected by competition,

herbivory, pollination, seed dispersal, and a range of climatic factors such as temperature, precipitation, etc. (Sakai et al., 1999).

In the tropics, few phenological studies have been carried out, most of these at the community level, including a number of arboreal species at a time. The phenology of other forms (i.e., shrubs and herbs) is relatively unknown, despite their importance for community diversity and structure (Madeira and Fernandes, 1999). The phenological characteristics of flowering (intensity, duration, and overlap) are important aspects of the reproductive effort of the plant. They have been quantified and related to reproductive success as measured by seed production (Guitián and Sánchez, 1992). Plants exhibit a variety of cyclic patterns in the aseasonal tropics, where favorable settings for flowering throughout the year result in a range of variation in timing of flowering (Bawa et al., 2003).

#### Gene dispersal/Reproductive strategies

For management and recovery programs to be successful, information on pollination biology and breeding systems of rare and endangered species is indispensable (Weller, 1994). Johnson and Sindel (2005) postulated that the reproductive strategy of a perennial weed was environmentally dependent. In their study sexual reproduction was favored when sites for establishment were relatively homogeneous and unfavorable and the number of potential establishment sites was small, while asexual reproduction was favored over seed when the environment was heterogeneous in terms of favorable and unfavorable establishment sites and the number of establishment sites was high.

## **Breeding systems**

Although the family Bignoniaceae includes approximately 800 species distributed in 112 genera, only 30 species have been investigated for their breeding systems (Bittencourt and Semir, 2004). In a broad sense, plant breeding systems are represented by the sum of sexual characteristics that directly influence the genetic composition of subsequent generations. The amount and distribution of genetic variation within populations can be dramatically influenced by breeding system characteristics, such as flower phenology, self-compatibility, and mating system (Kittlelson and Maro, 2000). As an example, one could postulate that genetic subdivision could result from high rates of autofertility, self-pollination, or restricted pollinator movement. Population subdivision can be prevented with self-incompatibility or even a low frequency of outcrossing, especially when pollen carryover is high or distant matings occur (Kittlelson and Maro, 2000).

The genetic architecture of a population can be interpreted by determining the proportion of outcrossed and selfed progeny and the level of inbreeding depression. In a population, the proportion of outcrossing and selfing depends on self-incompatibility mechanisms, floral development, and pollinator behavior. In self-compatible species, the probability of outcrossing increases with temporal separation of the male and the female reproductive phases. Autonomous selfing may occur frequently if pollen is proximal to the stigma and the stigma is receptive when the pollen is viable. Facilitated selfing may be high in plants with many inflorescences and where both female and male phases are mature because pollinators may forage longer among flowers of the same plant (Kittlelson and Maro, 2000).

According to Jesson and Barrett (2005), two important features of the biology of flowers influence their function. First, most plants produce hermaphroditic flowers and therefore reproduce as both female and male parents. Second, offspring arising from cross-fertilization are generally fitter than those from self-fertilization. The interpretation that most floral mechanisms function to promote cross-pollination comes as a result of the fitness consequences of cross- and self-fertilization.

### Pollination

Insects are the most common flower visitors in temperate zones; they have short lives and reproductive cycles that are synchronized with seasonal changes in climate and resource availability. In the climatically more uniform tropic zones, larger animals that are long-lived and require a year-round food supply are able to participate in this partnership (Tschapka and Dressler, 2002).

Bats are well-known as pollinators of various plant species in the Old and New World, e.g. the Phyllostomidae (Microchiroptera) in the Western Hemisphere and the Macroglossinae (Pteropidae: Megachiroptera) in Southeast Asia (Lack, 1978). Flowers have the ability to attract bats over different scales of distances. An example of important long-distance attractants are floral scents. Odors that humans frequently perceive as unpleasant and have been compared, for example, to over-ripe beans, sour milk, chlorine, mouse urine, human excrement, garlic or a cadaver are what characterize most bat-flowers. Scent is probably no longer the main guide and gets replaced by other cues once a bat approaches within a couple of meters of the flower. Contrary to popular belief, bats are not blind (Rodríguez-Durán, 2005; Tschapka and Dressler, 2002) and may also rely on visual cues to find flowers, especially in more open habitats like deserts (Tschapka and Dressler, 2002).

In Puerto Rico there are 13 species of bats today; in the past there were three more species that now are extinct (Rodríguez-Durán, 2005). These 13 species are distributed in five families: Noctilionidae, Mormoopidae, Phyllostomidae, Vespertilionidae and Molossidae. Of these 13 species, ten prefer caves as their shelter, two can be found in trees all day, and one prefers the roofs of wood houses and cracks in concrete structures (Rodríguez-Durán, 2005). Bats fly in groups of hundreds and thousands several kilometers each night in search of food; in Puerto Rico they feed on insects, fruits, flower nectar, leaves and fish. The bats that feed on fruits and nectar are responsible for seed dispersal and the pollination of flowers (Rodríguez-Durán, 2005). There are two species of bats in Puerto Rico that inhabit caves and visit flowers, Erophylla sezekorni (E. bombifrons), the "brown flower bat," and Monophyllus redmani, the "Greater Antillean long-tongued bat." Both species belong to the family Phyllostomidae, and both have been observed in the Maricao Forest Reserve (A. Rodríguez-Durán, pers. comm.). They have similar feeding habits, but *E. sezekorni* is mainly frugivorous, and eats fewer insects than *M. redmani*. It has been documented that *M. redmani* visits the flowers of columnar cacti, Furcraea, Ceiba, Musa, some palms, and Piper aduncum in Puerto Rico (Rodríguez-Durán, 2005). Erophylla sezekorni does not possess the capacity to hover in mid-air, so it has to land on the plant and would be expected to leave some kind of a mark or scar on the flower (A. Rodríguez-Durán, pers. comm.).

#### Hybridization

Raven et al. (2005) define a hybrid as the progeny of two parents that vary in one or more heritable characteristics, such as the progeny of two different varieties or species. One or more hybrids have been recorded for about 6 to 16% of plant genera, but hybridization is unevenly distributed across taxonomic groups (Mráz et al., 2005). To recognize a hybrid in the field, it is necessary to look at morphological characters, but according to Mráz et al. (2005) their use is limited mainly to cases of polyploid species complexes or introgression. In recent years molecular approaches (allozymes, DNA analysis) have resulted in important insights into the role of hybridization and plant speciation (Mráz et al., 2005). A main factor that promotes the creation of hybrids is species dispersal promoted by humans. Hybrids increase their success of spreading by overcoming natural crossing barriers, when disturbances such as fragmentation of habitats occur (Mráz et al., 2005; Rieseberg, 1991; Vilà et al., 2000). Among the factors considered critical for the creation of hybrids are the differences in the ecological predilection of parent plants (Mráz et al., 2005). In a study by Mráz et al. (2005) the parent plants of hybrids were separated by altitudinal and ecological demands (for example, light intensity). Hybrid plants became established at intermediate elevations in biotopes disturbed by human activities, where both parental species came into secondary contact.

According to Rieseberg (1991), hybridization could result in greater genetic diversity, increased fitness, and adaptation to new environments; in certain rare cases, hybridization may well be the only alternative to protect the germplasm of a rare or endangered taxon. The main harmful genetic effect of hybrids on native species is the loss of both genetic diversity (through the genetic integration of a smaller population by a larger one) and locally adapted populations, particularly in the case of rare and threatened species (Rieseberg, 1991; Vilà et al., 2000). Small relict populations (for example small island endemics), when they come in contact with a more numerous or reproductively more successful species, could have a problem with genetic assimilation (Rieseberg, 1991). Plants in islands become vulnerable to hybridization and genetic assimilation due to limits imposed by the geography and the soil, which limit species ranges and

population size (Rieseberg, 1991; Vilà et al., 2000). Other factors that promote hybridization in island plants are the lack of chromosomal sterility barriers between closely related species, and the invasion and colonization by closely related exotics (Rieseberg, 1991; Vilà et al., 2000).

#### Fruit and seed dispersal

Raven et al. (2005) define an angiosperm fruit "as a mature ripened ovary (or group of ovaries), containing the seeds." Fruits are classified as fleshy (seeds imbedded in succulent tissues) or dry (seeds not imbedded in succulent tissues). Dry simple fruits are classified as dehiscent fruits or indehiscent fruits. In dehiscent fruits, the seeds are freed when the tissues of the mature ovary wall (the pericarp) break open; in indehiscent fruits, seeds stay in the fruit after the fruit falls from the plant (Raven et al., 2005).

When seeds are dispersed they break away from density-dependent mortality near parents, take possession of open habitats and colonize remote sites in which the species is not present or locate microsites critical for establishment, and assist in the preservation of forest diversity (Howe and Miriti, 2004). Dispersed seeds have a higher chance of surviving to reproductive age than undispersed seeds. Small seeds have a higher chance of being dispersed and escaping direct competition with their maternal parent, but large seeds, if dispersed, produce large, well fed seedlings with better mechanisms to establish and live in the shaded understory (Howe and Miriti, 2004). A benefit of dispersal is escape from seed predators that exist near the parent plant or hunt for concentrations of seeds. Broadly scattered seeds have a lower chance of being destroyed or infested by predators than those in clustered masses (Howe and Miriti, 2004). By dispersing away from their parents or others of the same species (for example, if they land underneath a

plant of a different species rather than one of their own), seeds are saved from competition or from foci of pathogen or insect infestation (Howe and Miriti, 2004).

#### Agents of fruit and seed predation

When a seed is dispersed it suffers one of the following consequences: germination, entry into the seed bank (where it could lose its viability, suffer a fungal attack, or decompose), secondary dispersal, or consumption. A number of factors determine the likelihood that a seed will become prey before or after dispersal: the habitat into which the dispersed seed falls (habitat heterogeneity might lead to variation in the distribution of consumers), abundance of seed predators, and their foraging patterns and behavior (Reed et al., 2005). On the other hand if the seed falls into a patch of deep plant litter it might be difficult for predators to find, and the probability of survival could become greater; it could also worsen, if it looses viability, suffers a fungal attack, or decomposes.

# **METHODS**

### Distribution

For the purpose of this investigation, populations represent groups of individuals growing in the same locality. Location data for the populations were obtained in three ways. An interview was conducted with Mr. Rubén Padrón-Vélez, who had worked as a Department of Natural and Environmental Resources management official at the Maricao Forest Reserve. A search was done of the BRAHMS database at the MAPR Herbarium of the University of Puerto Rico, Mayagüez Campus, which gave information on specimens located at different herbaria in Puerto Rico and the United States (particularly MAPR, NY, and UPR). Location data were also obtained from the four reports submitted by Breckon and Kolterman under cooperative agreements number 14-16-0004-91-958, 14-16-0004-92-970, 14-16-0004-93-973 and 1448 0004-94-9113 between the U.S. Department of the Interior, Fish and Wildlife Service and the Department of Biology, University of Puerto Rico, Mayagüez Campus. All populations visited were mapped on USGS 7.5 minute topographic maps, and locations determined by GPS (Garmin etrex; 15 m circle of accuracy) were compared for accuracy with features shown on topographic maps. PR Datum was used when gathering GPS coordinates, DNER permit number 06-EPE-017 was assigned by the agency to the project.

## **Populations**

An extensive search was performed for new individuals (including seedlings) and populations in likely localities in the forests and in adjacent privately owned areas. Between December 2006 and February 2008 all 14 reported locations and two new locations (one of them found with the assistance of Mr. Adrián Muñiz-Suárez, former resident management official for the Maricao Forest Reserve) were extensively searched for individuals. GPS readings were obtained for individuals and populations. Habitat conditions (aspect, light conditions and associated species) were recorded for each population. Liogier and Martorell (2000) was consulted for authority and associated species family names. USGS topographic maps (7.5 minute series 1:20,000) were used in the determination of elevation, topographical features, and place names. U. S. G. S. Water Resources Data Puerto Rico and the U. S. Virgin Islands Water Year 1990 maps were consulted for drainage basin and river orientation. Ewel and Whitmore's (1973) map of Holdridge's Ecological Life Zones of Puerto Rico and the U. S. Virgin Islands was consulted to determine vegetation types. USDA Soil Survey Maps [Mayagüez Area, Puerto Rico Western Part (PR684) and San Germán Area, Southwestern Puerto Rico (PR787)] were used to determine the soil types; I-918 and (Llerandi-Román, 2004) maps were used to determine geological formations.

### **Field observations**

Initially, plants were labeled with numbered tags, but this practice was soon abandoned due to the fact that Breckon and Kolterman tagged almost all of the individuals throughout their four-year study. For all individuals basal diameter was measured with a caliper and the number of stems from the base was counted; the means for each population were calculated. For each plant, reproductive status (sterile, buds, flowers and/or fruits, including their size and color), and the number of cases of asexual reproduction were recorded. The number of cases per plant of recruitment by asexual (rooting of broken branches or of the terminal portions of the arching stems) and/or sexual reproduction in wild populations in the field was recorded. A study of the reproductive phenology (timing and duration of flowering) was conducted. Detailed observations of flowers (day and the time of day the flowers opened and color of the flower shown throughout its life) were conducted. Observations on flower visitors were recorded for ten minutes every half hour from late afternoon (19:00h) to early morning (7:00h). Fruits and plant characteristics based on Gentry's (1980) key [leaf size and shape, leaf surface (shiny vs. dull), fruit shape and size] were examined on all individuals found to assess the extent and impact of hybridization.

Information on fruits and seeds per fruit were gathered. Fruit color and measurement data were recorded in the field; fruits were then taken to a laboratory at the University of Puerto Rico, Mayagüez Campus, where they were opened and seeds were counted. Fruits found on the ground as well as on plants in the field were observed for dispersal, and examined for agents of fruit and seed predation.

### **Conservation status**

The conservation status of the species was evaluated, considering the species' recovery plan, and recommendations regarding its eventual recovery were made. Both U.S. Fish and Wildlife Service and the International Union for Conservation of Nature (IUCN) criteria were applied. For this purpose, the area of occupancy for each population was estimated using the Arc GIS 9 Version 9.2 computer program, the extent of occurrence for the species was estimated as described in Guidelines for Using the IUCN Red List Categories and Criteria Version 7.0, August 2008. Copies of the thesis will be submitted to the Department of Natural and Environmental Resources, the management officials, and the U.S. Fish and Wildlife Service.

# **RESULTS**

Between December 2006 to February 2008, a total of 42 field trips were conducted, 13 to the Maricao Forest Reserve and 29 to the Susúa Forest Reserve. *Crescentia portoricensis* was found in eight localities in the Maricao Forest Reserve and in three localities in the Susúa Forest Reserve (Figs. 11, 12 and 13). A total of 532 individuals were found. The number of main stems from the base and main stem diameter on April 3, 1993 and on January 11, 2008 for previously tagged individuals in the Río Maricao population, Maricao Forest Reserve are presented in Table 4, while the number of main stems from the base and main stem diameter for previously tagged individuals in the Quebrada Peces and Quebrada Grande populations and in the Arroyo del Tanque subpopulation, Susúa Forest Reserve are presented in Table 5. The extent of occurrence for the species was 43 km<sup>2</sup>.

In the remainder of this section, each of the populations in Maricao and Susúa is discussed, including information on locality, number of individuals and reproductive status. Asexual reproduction refers to rooting of arching stems and branches; the final stage of asexual reproduction was not observed during this study, as branches and stems were not observed to break off plants to form separate individuals.

# **Maricao Forest Reserve populations**

# **Population 1: Río Maricao**

The Río Maricao population was visited on January 11 and 13 of 2008. It is located in Bo. Maricao Afuera in the Municipality of Maricao, north of the Department of Natural and Environmental Resources Fish Hatchery (Fig. 14). The Río Maricao is part of the Río Guanajibo drainage basin and flows generally north and then east (Curtis et al., 1991). The population can be reached by walking upstream along the Río Maricao from the DNER Fish Hatchery. An alternate route to access the population, is by walking down the Viveros trail which on the east side of Rd. 120 between kms. 14.8 and 14.9. Plants can be found along both sides of Río Maricao. Some plants were as close as 0.5 m to the water's edge; the farthest plant was found 6.1 m from the water's edge. Thirty-eight plants were found in the Río Maricao population. Eighteen plants were found below the first fork. Above the fork, 14 plants were found along the eastern tributary and six plants along the western tributary. Only five plants were found in eight groups (defined as a cluster of plants of which none is more than four meters from another individual of the species): one group of two individuals, four groups of three individuals, one group of four individuals (located on flat ground by the river), one group of five individuals and one group of six individuals. The nearest population was the Quebrada Piedras population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). Four plants showed sexual reproduction, and four showed asexual reproduction. Three plants had flower buds; one of them had 10 flower buds, all on a branch oriented away from the closed forest and into the river. The plant #341 and an untagged plant had one flower bud each. The number of fruiting plants and the mean size of fruits per plant were calculated (Table 6). One plant had a dark green fruit that measured 9.0 cm x 2.8 cm. Roots were observed on a branch of an untagged plant that rested directly on the ground; this branch had not detached to form a new individual. On two plants, plant #325 and an untagged plant, roots were observed growing from the tip of a branch. On another untagged plant, a shoot was observed rising from a buried branch; excavation revealed that a root had been produced at the base of this shoot.

Site description:

Substrate: Caguabo clay loam, Rosario clay, serpentine outcrop and remnants of lateritic residual soil on serpentinite (Cretaceous).
Coordinates: 18° 09' 53.8" N, 066° 59' 21.5" W.
Elevation: 490 – 575 m.
Area of occupancy: 18,322 m<sup>2</sup>
Aspect: 58° NE
Light conditions: Open canopy, sparse, the plants were almost completely exposed.
Life Zone: Subtropical Wet Forest
Associated species: Arthrostylidium sp. (Poaceae), Clusia rosea (Clusiaceae).

## **Population 2: Quebrada Seca**

The Quebrada Seca population was visited on January 25, 2008. The population is located in the northwest of Bo. Tabonuco in the Municipality of Sabana Grande (Fig. 15). The Quebrada Seca is part of the Río Guanajibo drainage basin and flows from the northwest to the southeast (Curtis et al., 1991). The population can be reached by walking down (southeast) into the closed forest from Rd. 366 at Km 0.2. Plants can be found along both sides of the quebrada, which was dry at the time of the visit. Five plants were found in the quebrada. Only one plant (#305) was found to have a tag; the other four were untagged, and it was assumed that they had lost their tags. The plants were found in one group of one individual and two groups of two individuals. The nearest population was the Río Postrero population.

The means of the number of main stems from the base and stem diameters are presented in Table 2. Only three plants were found in reproduction; two showed sexual reproduction, and one showed asexual reproduction. One plant had a flower bud, a flower and a fruit; another plant had just one flower. From an untagged plant, roots were observed along two branches. The area could not be searched further because this section of the quebrada is at the top of a waterfall.

Site description:

Substrate: El Descanso-Hoconuco complex on serpentinite (Cretaceous).
Coordinates: 18° 08' 34.1" N, 066° 57' 25.8" W.
Elevation: 707 – 716 m
Area of occupancy: 55 m<sup>2</sup>
Aspect: 124° SE
Light conditions: Open canopy, sparse, the plants were almost completely exposed.
Life Zone: Subtropical Wet Forest
Associated species: *Simarouba tulae* (Simaroubaceae).

## **Population 3: Quebrada Piedras**

The Quebrada Piedras population was visited on January 8 and 9, 2008. It is located in the boundary between the Bo. Hoconuco Alto and Maricao Afuera in the Municipalities of San German and Maricao respectively (Fig. 16). The Quebrada Piedras is part of Río Guanajibo drainage basin and flows from the northeast to the southwest (Curtis et al., 1991). The population can be reached by walking up (northeast) the quebrada by the forest reserve manager's residence at the Maricao Forest Reserve. Nine wild plants were found in Quebrada Piedras; two plants can be found in the southwestern part quebrada before it crosses Rd. 120, and another seven individuals were found in the northeastern part of the quebrada near the telecommunications antennas. Sixty-four plants were planted by DNER personnel in the quebrada, 21 of these by the resident forest manager official's house and 43 upstream west of the old water tank. Water was not observed in any of the trips made to the quebrada, but employees of the DNER say that it becomes active in the rainy season. Seven of the plants in this population were tagged (#231, 273-278) on the first trip made to Quebrada Piedras; another two were left untagged because these were not observed in the first trip made to the quebrada. The plants were found in two groups of one individual, one group of three individuals, and one group of four individuals. The nearest population was the Quebrada Negra population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). No plants in the population were found in a reproductive state.

Site description:

**Substrate:** El Descanso-Hoconuco complex, Nipe clay, remnants of lateritic residual soil on serpentinite (Cretaceous).

**Coordinates:** 18° 09' 10.0" N, 066° 59' 28.4" W.

**Elevation:** 804 – 834 m

Area of occupancy: 40 m<sup>2</sup>

Aspect: 39° NE

Light conditions: Open canopy, sparse, the plants were almost completely exposed. Life Zone: Subtropical Wet Forest

Associated species: Arthrostylidium sp. (Poaceae), Buchenavia tetraphylla (Combretaceae), Prestoea montana (Arecaceae).

#### **Population 4: Río Bonelli**

The Río Bonelli population (a previously unreported group) was visited on January 16, 2008. The population is located on the boundary of the Bo. Maricao Afuera and Indiera Fría in the Municipality of Maricao (Fig. 17).

The previously reported individual (Breckon and Kolterman, 1992) on a "moist, steep, Nfacing slope on the N side of Hwy. 120 at Km 14.0, across from the Observation Tower" was searched for, but was not found. The Río Bonelli is part of Río Grande de Añasco drainage basin and flows from the south to the northeast where it joins Río Lajas (Curtis et al., 1991). The population can be reached by following the dirt road at the end of Rd. 425. This dirt road will intersect Río Lajas first and Río Bonelli second; the population can be found upstream from that second intersection. Plants can be found along both sides of Río Bonelli. Plants were as close as 2.2 m and as far as 17.2 m from the water's edge. Sixteen plants were found in the Río Bonelli. The plants were found in two groups of one individual, two groups of two individuals, and one group of ten individuals. The first group found at the river was a group of two plants, 18 m from there in a small valley (east) beside the river an individual was found, and further north at an unmeasured distance another individual was found. A group of ten individuals was found at a distance of 210 m after the second solitary plant. The last group (group of 2) found at the river was 14 m from the group of ten. The nearest populations were the Río Postrero and Río Lajas populations, which were found to be at the same distance from the Río Bonelli population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). Eight plants were found in reproduction: four showed sexual reproduction, and four showed asexual reproduction. One plant had seven flower buds and a flower, another plant had a bud and a flower, and another plant had three buds. The number of fruiting plants and

the mean size of fruits per plant were calculated (Table 6). A plant was found to have two dark green fruits, which were collected to count their seeds. Information on fruit color and size and seeds per fruit was recorded (Appendix III). Roots were observed along four branches on four different individuals; these branches rested directly on the ground. On a branch of one of these individuals a shoot rose up from the ground; on a branch of another individual two shoots were observed.

Site description:

Substrate: Caguabo clay loam and Humatas clay on the Río Loco formation of Slodowski (1956) and Mattson (1960) (Upper Cretaceous) and serpentinite (Cretaceous)
Coordinates: 18° 09' 57.9" N, 066° 58' 04.7" W.

**Elevation:** 273 – 448 m

Area of occupancy: 1,323 m<sup>2</sup>

Aspect: 119° SE

Light conditions: Open canopy, sparse, the plants were almost completely exposed.

Life Zone: Subtropical Wet Forest

Associated species: Arthrostylidium sp. (Poaceae), Clusia rosea (Clusiaceae).

## **Population 5: Río Cupeyes**

The Río Cupeyes population (a previously unreported group) was visited on February 3, 2008. The population is located in the Bo. Santana, in the Municipality of Sabana Grande (Fig. 18). The Río Cupeyes is part of the Río Guanajibo drainage basin and flows from the north to the southwest (Curtis et al., 1991). The Río Cupeyes population was reached by walking down old

dirt Rd. 362 (south of Campamento Santana); the plants can be found west of the dirt road in a quebrada that flows under a cement bridge. The first plant in the population was found at a distance of 47 m from the bridge. Water was observed at the time of the visit; the plants were as close as 4.04 m and as far as 5.3 m from the water's edge. Twenty-seven plants were found in this population. The plants were found in a group of one, a group of five, a group of seven, and a group of 14. The nearest population was the Quebrada Negra population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). Seven plants were found in reproduction: four showed sexual reproduction, and three showed asexual reproduction. One plant had a flower bud; two plants had a bud and a flower. The number of fruiting plants and mean size of fruits per plant were calculated (Table 6). A plant showed a bud, a flower and a dry dark brown fruit that measured 8.7 cm x 4 cm. Two other plants showed roots at the tip of one branch each; a branch of another individual rested on the ground and roots were observed growing along it.

Site description:

Substrate: El Descanso-Hoconuco complex on serpentinite (Cretaceous) Coordinates: 18° 08' 16.4" N, 066° 58' 19.3" W. Elevation: 678 – 690 m Area of occupancy: 165 m<sup>2</sup> Aspect: 116° SE Light conditions: Open canopy, sparse, the plants were almost completely exposed. Life Zone: Subtropical Wet Forest Associated species: *Cecropia schreberiana* (Moraceae), *Clusia rosea* (Clusiaceae), *Comocladia glabra* (Anacardiaceae), *Simarouba tulae* Urb. (Simaroubaceae).

### **Population 6: Río Postrero**

The Río Postrero population was visited on January 14, 2008. The population is located in the Bo. Indiera Fría, in the Municipality of Maricao (Fig. 19). The Río Postrero is part of the Río Grande de Añasco drainage basin, it flows from the southeast to the north, where it joins Río Lajas (Curtis et al., 1991). The Río Postrero population was found by ascending the drainage on the north side of Rd. 120 between Kms. 11.7 and 11.8. The first plant in the population was found at a distance of 16.6 m from the road. The river was dry, and only two plants were found at the site. The nearest population was the Quebrada Seca population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). Two plants were found in reproduction; one showed sexual reproduction and the other showed sexual and asexual reproduction. The two plants were found to have one bud while one of them showed roots growing at the tips of two of its branches. These branches were observed to have crossed from one side of the drainage to the other.

Site description:

Substrate: El Descanso-Hoconuco complex on serpentinite (Cretaceous) Coordinates: 18° 08' 50.5" N, 066° 57' 46.4" W. Elevation: 730 m Area of occupancy: 10 m<sup>2</sup> Aspect: 135° SE Light conditions: Open canopy, sparse, the plants were almost completely exposed. Life Zone: Subtropical Wet Forest

Associated species: Cecropia schreberiana (Moraceae), Clusia rosea (Clusiaceae), Comocladia glabra (Anacardiaceae), Simarouba tulae (Simaroubaceae).

## **Population 7: Río Lajas**

The Río Lajas population was visited on January 14, 2008. The population is located in the Bo. Indiera Fría in the Municipality of Maricao (Fig. 20). The Río Lajas is part of Río Grande de Añasco drainage basin and flows from the southeast to the northwest, connecting with Río Bonelli. The tributary in which the plants are found meets the Río Lajas from the southeast; it drains into the river below Salto Curet. The population can be most easily reached by walking along a bulldozed dirt road that is parallel to (east of) the Río Lajas. At the time of the visit, the tributary crossed and flowed on top of the bulldozed dirt road. Plants were found along both sides of the tributary. Plants were as close as 1.3 m and as far as 3.9 m from the water's edge. Nine plants were found in the tributary. One of the plants was found to be cut with a machete, maybe to make space for the plastic container and the PVC tubing that collected water from the tributary and discharged it into Río Lajas (Fig. 21). The plants were found in a group of two individuals, and a group of seven individuals. The first group found at the tributary was the group of seven which is 42 m from the road, while the group of two was found at a distance of 15 m from the group of seven. The nearest population were the Río Bonelli and Río Postrero which were found to be at the same distance from the Río Lajas population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). None of the plants was found in a reproductive state.

#### Site description:

Substrate: Serpentine outcrop on the Río Loco formation of Slodowski (1956) and Mattson (1960) (upper Cretaceous). Coordinates: 18° 09' 59.7" N, 066° 57' 41.7" W. Elevation: 414 m Area of occupancy: 10 m<sup>2</sup> Aspect: 98° SE

Light conditions: Open canopy, sparse, the plants were almost completely exposed. Life Zone: Subtropical Wet Forest

# **Population 8: Quebrada Negra**

The Quebrada Negra population, a previously unreported population, was visited from January 25 to 27 of 2008. It is located in Bo. Maricao Afuera and Bo. Santana in the Municipalities of Maricao and Sabana Grande, respectively (Fig. 22). Quebrada Negra does not appear on the topographic map, but it appears to be part of the Río Grande de Añasco basin and flows north to Río Bonelli. The plants grow on flat ground. The population can be accessed by entering the closed forest into the recreational area called "La Caballeriza", which is on the north side of Rd. 120 between Kms. 12.9 and 13.0, 38 m west of the "Estación de Bomba Relevo" of the Puerto Rico Aqueduct and Sewer Authority. "La Caballeriza" recreational park was inaugurated in 1968 under the administration of Luis A. Ferré; the area was managed at that time by the "Compañia de Fomento Recreativo". The area was supposed to become a horse riding park; stables were built but a proposed motel was never constructed, and the project was soon abandoned due to the area's rocky surface. The site was used as a passive recreational area

(merenderos); there are still some gazebos and picnic benches. The area was later abandoned; the riding trails were reopened in 2003 by the "Compañia de Parques Nacionales" and later in 2007 by DNER employees. This might explain why one plant was cut with a machete. The "Compañia de Parques Nacionales" does not have any plans to develop the area for now, but they plan to build some villas in the old parking lot of the stables at Km 13.2. According to Mr. Padrón-Vélez the population is a combination of wild plants and an unknown number of planted individuals. Plants can be found along both sides of Quebrada Negra. Plants were as close as 0.5 m and as far as found 10.9 m from the water's edge. Fifty-seven plants were found in this population. The nearest population was the Río Cupeyes population.

The means of the number of main stems from the base and stem diameters were calculated (Table 2). Only two plants were found in asexual reproduction. On one plant a stem 2.08 cm in diameter was almost completely buried, and roots grew along it. On another individual a 2.21 cm stem rested on the ground and two groups of roots were observed growing from it.

# Site description:

Substrate: Rosario clay, El Descanso-Hoconuco complex, Cerro Gordo mucky peat and remnants of lateritic residual soil on serpentinite (Cretaceous)

**Coordinates:** 18° 08' 29.6" N, 066° 58' 18.2" W.

**Elevation:** 814 – 818 m

Area of occupancy: 28 m<sup>2</sup>

Light conditions: Open canopy, sparse, the plants were almost completely exposed. Life Zone: Subtropical Wet Forest Associated species: Arthrostylidium sp. (Poaceae).

# Susúa Forest Reserve populations

#### **Population 9: Quebrada Peces**

The Quebrada Peces population was visited on eight trips in 2007: March 11, 16, 18, and 22, April 20, June 27, and August 23 and 26. The population is located in the Barrios Torre and Susúa Alta, on the boundary between the Municipalities of Sabana Grande and Yauco, respectively (Fig. 23). The Quebrada Peces is part of the Río Loco drainage basin and flows from the northwest to the southeast, where it joins the Río Cañas (Curtis et al., 1991). The population can be reached by entering the quebrada from the road that gives the public access to the DNER offices. Plants were found along both sides of Quebrada Peces, and were as close as 0.2 m and as far as 12 m from the water's edge. One hundred seventy plants were found in Quebrada Peces: 113 were found north of the road, while 57 were found south of the road. Also, south of the road a plant of *Crescentia cujete* tagged as #240 was growing by the river. Figure 24 shows the distribution of plants along Quebrada Peces and its tributaries.

Forty-nine plants were found with tags; the numbered tags found in the north part of the population were #101, 103, 105-111, 113-117, 119-121 and 200. Numbered tags in the south part of the population were #123-125, 127-131, 133-140, 142, 143, 145-150, 152-157, and 160. In this population the plants were found in nine groups of one individual, two groups of two individuals, four groups of three individuals, three groups of four individuals, two groups of five individuals, two groups of seven individuals, one group of nine individuals, and one group of 28 individuals. The nearest population was the Río Loco population.

The means of the number of main stems from the base and stem diameters were calculated (Table 3). A total of 34 plants were found in reproduction, 19 showed sexual reproduction, 12 showed asexual reproduction, and three plants showed both sexual and asexual reproduction. The number of fruiting plants and the mean size of fruits per plant were calculated (Table 6). A total of 55 fruits were observed; 23 of these were unreachable and were not measured. Flower buds were only found in one plant which produced two; this plant had a green fruit also. Under plant #103 part of an old pericarp was found on the ground, seeds and termites were observed in it. Termites were not observed carrying or dispersing the seeds, it was inferred that they were feeding on the seeds. One of the plants in the south group was observed to have three fruits growing on the same branch (Fig. 24). Roots were observed at the tip of a branch of six individuals; four of these had been tagged (#116, 121, 142, and 160). On a slope behind plant #117, an untagged plant had a small branch (0.64 cm) connected to two stems (Fig. 25). This branch had not detached to form a new individual. Roots were observed along a buried branch of three individuals (#129 and two untagged plants); from the two untagged individuals a shoot was observed arising from the buried branch. On plant #134 a stem was observed to bifurcate and roots were seen along the bifurcations. Only in plant #120 a branch was observed to have detached to form a new individual. An untagged plant was observed to have one dark green fruit which measured 9.6 cm x 2.3 cm and a 2.74 m branch that connected to what seemed another adult plant; a second plant was observed to be connected underground by a root. From a branch (that rested on the ground) of the same plant, two new plants were observed to have emerged from two pieces of the branch that had detached, and two other pieces had not yet detached to form new individuals. Plant #133 was observed to have a green fruit 7.2 cm x 2.5 cm and, at the tip of a branch that rested on the ground, roots were observed. An untagged plant was observed

to have five fruits, one black that measured 11.3 cm x 3.1 cm, one greenish brown that measured 9 cm x 2.6 cm, and three dark green that measured 9.7 cm x 2.5 cm, 9.3 cm x 2.6 cm and 10.9 cm x 2.7 cm. Roots were also observed at a tip of a branch on this plant. These roots were only observed when the branches touched the ground.

Site description:

**Substrate:** El Cacique-La Taína complex and Quebrada clay loam on serpentinite (Cretaceous).

**Coordinates:** 18° 04' 29.3" N, 066° 54' 45.4" W (north section of the quebrada), 18° 04' 16.6" N, 066° 54' 36.7" W (south section of the quebrada).

**Elevation:** 158 – 292 m

**Area of occupancy:** 57,769 m<sup>2</sup>

Aspect: 47° NE

Light conditions: Under thick canopy, the plants were not exposed.

Life Zone: Subtropical Moist Forest

Associated species: Clusia rosea (Clusiaceae), Mangifera indica (Anacardiaceae), Ottoschulzia rhodoxylon (Icacinaceae), Pimenta racemosa (Myrtaceae), Tabebuia haemantha (Bignoniaceae).

# **Population 10: Quebrada Grande**

A known subpopulation (subpopulation 1) of the Quebrada Grande population was visited on January 7, 2008, and a previously unreported subpopulation was discovered on November 13, 2007. Quebrada Grande divides the barrios Susúa Alta (west) and Collores (east)

in subpopulation 1 and divides the barrios Ranchera (west) and Collores (east) in subpopulation 2. All of these barrios are located in the Municipality of Yauco. The Quebrada Grande is part of the Río Loco drainage basin and flows from the northwest to the southeast where it, as well as Quebrada Fría, join the Río Loco before it drains into the Presada Loco (Curtis et al., 1991). For subpopulation 1 (Fig. 26), all of the plants located to the north, and the plants located to the south that are on the east side of the quebrada, are on the Susúa Forest Reserve. The plants located to the south that are on the west side of the quebrada are on private lands. For subpopulation 2 (Fig. 27), the west side of the quebrada is in the Susúa Forest Reserve, while the east side (where the plants where found) is on private property. Subpopulation 1 can be reached by walking ca. 2 km southeast from the DNER offices along a dirt road that runs more or less parallel to the Río Loco. At the end of the dirt road (which becomes narrow), Quebrada Grande meets Río Loco; from there, Quebrada Grande can be walked upstream. The previously unreported subpopulation 2, which is the northern group of the population, can be accessed by hiking along Camino Elión A. Rancheras (west of Quebrada Grande) that is parallel to the quebrada; from there one must hike eastward down the Camino al Lago. The Camino al Lago ends at the point where a population of Calliandra locoënsis begins; after that one must walk through the closed forest to gain access to Quebrada Grande. In subpopulation 2, the plants were not found to be growing on a slope, but in a small valley by the river. The plants were as close as 0.7 m and as far as 10.4 m from the water's edge. Thirty-nine plants were found in the Quebrada Grande, 35 in subpopulation 1 and four in subpopulation 2. Only two plants in subpopulation 1 were found with tags; the numbered tags found were #204 and #216. The plants were found in eight groups of one plant, three groups of three plants, two groups of four plants (one of them in the previously unreported subpopulation 2), one group of six plants and one group of eight plants. The nearest population

was the Río Loco population. The means of the number of main stems from the base and stem diameters were calculated (Table 3).

A total of five plants were found in reproduction; one showed sexual reproduction, and four showed asexual reproduction; no plants were found in reproduction in subpopulation 2. The number of fruiting plants and mean size of fruits per plant were calculated (Table 6). One untagged plant had two dark green fruits, which measured 10.5 cm x 3.1 cm, and 12.0 cm x 3.0 cm. An untagged plant had a stem that rested on the ground and roots were observed along it. On another untagged plant roots were observed at the tip of branch that rested on the ground. On an untagged plant, one of the stems made contact with the ground and roots were observed at the tip of a branch; on another plant two stems lay on the ground and roots were observed along them.

Site description:

**Substrate:** El Cacique-La Taína complex and Maresúa serpentine outcrop complex on serpentinite (Cretaceous).

Coordinates: 18° 04' 36.2" N, 066° 53' 50.4" W (subpopulation 1), 18° 05' 30.6" N,

066° 53' 45.5" W (subpopulation 2).

**Elevation:** 134 – 170 m (subpopulation 1), 227 m (subpopulation 2)

**Area of occupancy:** 7,838 m<sup>2</sup>

Aspect: 139° SE (subpopulation 1)

Light conditions: Under thick canopy, the plants were not exposed.

Life Zone: Subtropical Moist Forest

Associated species: Calophyllum calaba (Clusiaceae), Clusia rosea (Clusiaceae), Pimenta racemosa (Myrtaceae).

### **Population 11: Río Loco**

The Río Loco population is divided into five subpopulations. Four of the subpopulations are along tributaries of the river, and a previously unreported subpopulation is located on the river itself. By the forest manager's house, right before one starts to descend along the Camino al Río a plant of *Crescentia alata* was found growing; the plant is identified as *Crescentia portoricensis* by a carved piece of wood that rests on the ground.

## Subpopulation 1: Quebrada Calliandra

The Quebrada Calliandra (name does not appear on the topographic map; cf. García and Kolterman, 1992) subpopulation was visited on December 24, 2007; it is located in the Bo. Susúa Alta, in the Municipality of Yauco (Fig. 28). The Quebrada Calliandra is part of the Río Loco drainage basin and flows from the northwest into the Río Loco (Curtis et al., 1991). This subpopulation can be reached by following the trail parallel to the Río Loco. The Camino al Río starts by the DNER resident management official's residence, where one would cross the river for the first time. One would follow the Camino al Río and cross the river two more times. At the third cross (called Charco La Mesa), one walks upstream along Río Loco; Quebrada Calliandra is the second tributary joining the Río Loco from the west. Plants were found along both sides of the quebrada; they were as close as 2.17 m and as far as 8.2 m from the water's edge. Fourteen plants were found at the site; of these, only two had tags (#162 and #165). In this subpopulation plants were found in a group of one, a group of six, and a group of seven individuals. The first cluster found was the cluster of six, which is at a distance of 119 m from the cluster of seven. The solitary individual found at a distance of 101 m from the second cluster. The nearest

population was the Quebrada Peces population. The means of the number of main stems from the base and stem diameters were calculated (Table 3).

Only two plants were observed in asexual reproduction. On an untagged plant two branches were observed to come in contact with the ground; roots were observed at their tips but these branches had not detached to form new plants. On another untagged individual, a branch rested on the ground and roots were observed along it; two shoots were observed rising from the ground, but these had not detached.

Site description:

Substrate: El Cacique-La Taína complex on serpentinite (Cretaceous)
Coordinates: 18° 05' 29.0" N, 066° 54' 40.8" W.
Elevation: 136 – 222 m
Area of occupancy: 137 m<sup>2</sup>
Aspect: 106° SE
Light conditions: Under thick canopy; the plants were not exposed.
Life Zone: Subtropical Moist Forest
Associated species: Calliandra locoënsis (Mimosaceae), Clusia rosea (Clusiaceae), Comocladia glabra (Anacardiaceae), Pimenta racemosa (Myrtaceae), Simarouba tulae (Simaroubaceae), Tabebuia haemantha (Bignoniaceae).

## **Subpopulation 2:**

Subpopulation 2 was visited on January 5, 2008; it is located in the Bo. Susúa Alta, in the Municipality of Yauco (Fig. 29). The subpopulation 2 is in a quebrada that flows from the north

and drains in the Río Loco. This group can be reached by following the Camino al Río; after the second cross of the river, subpopulation 2 is found in the third quebrada that intersects with the trail. Plants were found along both sides of the quebrada, and they were as close as 0.3 m and as far as 10.8 m from the water's edge. Forty-one plants were found in this quebrada. Only sixteen were tagged; these were #168-177, 179, 180, 182, 184, 185, 187 and 189. The nearest population was the Quebrada Peces population. The means of the number of main stems from the base and stem diameters were calculated (Table 3).

Only three plants were observed in asexual reproduction. Plant #172 showed roots more or less at the middle of a branch. On plant #179, at two of its branch tips and at more or less the middle of four other branches roots were observed; on one of these branches, roots were also at the end. On plant #182 roots were observed at more or less the middle of a branch.

Site description:

Substrate: El Cacique-La Taína complex on serpentinite (Cretaceous)
Coordinates: 18° 05' 30.0" N, 066° 54' 33.4" W.
Elevation: 281 m
Area of occupancy:
Aspect: 93° SW
Light conditions: Under thick canopy, the plants were not exposed.
Life Zone: Subtropical Moist Forest
Associated species: Comocladia glabra (Anacardiaceae), Pimenta racemosa (Myrtaceae).

## **Subpopulation 3**

Subpopulation 3 was visited on January 4, 2008; it is located in Bo. Susúa Alta, in the Municipality of Yauco (Fig. 29). The subpopulation is in a quebrada that flows from the north until it meets the Río Loco. This subpopulation can be reached by following the trail that runs parallel to the Río Loco; after the second cross of the river, the group will be in the second quebrada that intersects with the trail. Plants were found along both sides of the quebrada; they were as close as 0.8 m and as far as 4.52 m from the water's edge. Four plants were found upstream, north of the trail, of which three had tags (#191, 195 and 196). Fifteen plants were found south of the trail, of which three had tags (#197-199). The last individual found in the south group was 77.4 m from the trail. The nearest population was the Quebrada Peces population. The means of the number of main stems from the base and stem diameters were calculated (Table 3).

The number of fruiting plants and the mean size of fruits per plant were calculated (Table 6). Two plants were found with fruits, an untagged plant in the north, which had a dark green fruit that measured 9.4 cm x 3.8 cm, and #199, which had two dark green fruits that measured 9.6 cm x 2.8 cm and 10.0 cm x 3.0 cm. Three plants (untagged) were observed in asexual reproduction, in the south portion of the population. On one of them, roots were observed in three different places along a branch. On another plant, a branch reached across the quebrada; roots were observed at the tip of it and a new individual was emerging but had not detached. On another plant, at the tip of a branch roots were observed and a new individual that had not detached was emerging.

Site description:

Substrate: El Cacique-La Taína complex on serpentinite (Cretaceous)
Coordinates: 18° 05' 30.0" N, 066° 54' 33.4" W.
Elevation: 281 m
Area of occupancy:
Aspect: 24° NE
Light conditions: Under thick canopy, the plants were not exposed.
Life Zone: Subtropical Moist Forest
Associated species: Comocladia glabra (Anacardiaceae), Pimenta racemosa (Myrtaceae).

## Subpopulation 4: Arroyo del Tanque

The Arroyo del Tanque group was visited on September 7 and 13 of 2007; the group is located north of the Susúa Forest Reserve DNER offices in the Bo. Susúa Alta in the Municipality of Yauco (Fig. 30). Arroyo del Tanque is part of the Río Loco drainage basin and flows from the northeast until it meets the Río Loco. The population can be reached by walking along the Camino al Río. After the first cross of the river, one must stay on the Camino al Río (parallel to Río Loco); a cement bridge crosses a creek, and Arroyo del Tanque flows underneath that bridge. The arroyo was given that name because of the water tank in it (W. Cordero, pers. comm.). Plants were found along both sides of the arroyo. Some plants were as close as 0.5 m and as far as 11.5 m from the water's edge. Seventy-one plants were found in this population; six of these 71 grow along a small tributary that joins the arroyo from the north, while the remaining 65 plants grow in the slopes of the arroyo. Forty-nine plants were found with tags. The numbered

tags were #4, 6, 8-12, 14, 15, 17, 18, 21-23, 25-29, 35, 36, 38-46, 48, 50, 54-58, 63-65, 67, 68, 70-72, 91, 93, 95, 96; tags #44-46 were found along the tributary of the arroyo. Plants #33 and #34 were dead but still had tags on them. Plant #93 was found to be cut almost to the ground; its stems and branches were neatly arranged beside what was left of the plant. Apparently this plant was cut by DNER employees while repairing the pipes that collect water from the arroyo and supply it to the DNER offices (Fig. 31). Plant #93 was visited on December 6, 2008 and was found to be dead; so was plant #35, and tag #37 was found in the arroyo. In September of 1991 Breckon and Kolterman also found the trunk of plant #22 to be cut. The nearest population was the Quebrada Peces population. The means of the number of main stems from the base and stem diameters were calculated (Table 3).

Sexual reproduction was not observed in any of the plants in the arroyo group; asexual reproduction was observed on eight plants. On plant #23, on two branches that rested on the ground, roots were observed at the tips; the individuals had not detached. Roots were also observed along the stems of plants #40 and #71; the stem on #40 was buried. Plant #39 and an untagged plant showed roots on a branch. The branch on plant #39 was completely buried; the branch of the untagged plant lay on the ground. Plants #25 and #71 had roots along two branches; on plant #71 these branches were on two different main stems.

Site description:

Substrate: El Cacique-La Taína complex on serpentinite (Cretaceous) Coordinates: 18° 04' 28.6" N, 066° 54' 17.4" W. Elevation: 137 – 234 m Area of occupancy: 224 m<sup>2</sup> Aspect: 320° NW

Light conditions: Under thick canopy; the plants were not exposed.

Life Zone: Subtropical Moist Forest

Associated species: Clusia rosea (Clusiaceae), Comocladia glabra (Anacardiaceae), Pimenta racemosa (Myrtaceae), Randia aculeata (Rubiaceae), Simarouba tulae (Simaroubaceae), Tabebuia haemantha (Bignoniaceae).

## **Subpopulation 5**

Subpopulation 5 of the Río Loco population was visited on October 17, 2007 and from 18 to 30 December, 2007. It is located north of the Susúa Forest Reserve DNER offices in the Bo. Susúa Alta in the Municipality of Yauco (Fig. 32). The Río Loco is part of the Río Loco drainage basin and flows from the north to the southeast where it is joined by Quebrada Grande before draining into Presada Loco (Curtis et al., 1991). The subpopulation can be reached by walking along the Camino al Río; then one must enter the Arroyo del Tanque at the cement bridge and follow the arroyo until it drains into the Río Loco. From where the arroyo drains, the Río Loco can be walked upstream. All of the plants were found along the east side of the river, this being the only subpopulation of the Río Loco actually on the river itself. Río Loco was searched for plants, from the DNER offices (south) to where the river is formed by two quebradas, north on the boundary between the barrios Frailes and Ranchera. This is where one plant of Crescentia cujete was found. Plants were as close as 6.4 m and as far as 10 m from the water's edge. Fourteen plants were found in this population, 11 in the south group, which were the ones closest to the DNER offices, and three more in the north group, at a distance of 97.2 m from the southern group. No plants were found with tags, since this was a previously unreported group.

The nearest population was the Quebrada Peces population. The means of the number of main stems from the base and stem diameters were calculated (Table 3).

Sexual reproduction was observed on four plants. The number of fruiting plants and the mean size of fruits per plant were calculated (Table 5). In the southern group a plant was observed to have two flower buds; another individual had a yellow-orange fruit that measured 10.6 cm x 3.1 cm. In a plant in the northern group a bud and two dark green fruits, which measured 7.6 cm x 2.7 cm and 8.0 cm x 2.1 cm, were observed. On another plant in the same group a flower was observed; this flower showed a slit at its base. Asexual reproduction was observed on three plants, two in the northern and one in the southern group. On a plant in the northern group roots were observed more or less at the middle of a branch, and far down toward the end of that same branch. On another plant, at the tip of a branch, roots and a shoot were observed. On a plant in the southern group, roots were observed along a stem that rested on the ground, and on a branch of that stem that also rested on the ground. Two flowers on a plant in the southern group were observed for flower longevity for 13 days from December 18 to 30, 2007, mostly from 8:30 to 9:30 AM (Appendix IV).

Site description:

Substrate: El Cacique-La Taína complex on serpentinite (Cretaceous) Coordinates: 18° 04' 39.2" N, 066° 54' 25.0" W. Elevation: 127 – 145 m Area of occupancy: 16 m<sup>2</sup> Aspect: 290° NW

Light conditions: Under thick canopy the plants were not exposed.

# Life Zone: Subtropical Moist Forest

Associated species: Clusia rosea (Clusiaceae), Mangifera indica (Anacardiaceae), Ottoschulzia rhodoxylon (Icacinaceae).

# DISCUSSION

# Localities

GPS readings were not obtained for some of the plants, because the GPS receiver used in the study could not receive the satellites' signals. Interference of the signal might have been caused by local topography, closed canopy or cloud cover. These localities were in the Río Maricao, Quebrada Piedras, Río Bonelli, Río Cupeyes, Río Postrero and Río Lajas populations in the Maricao Forest Reserve and Quebrada Peces, subpopulation 1 of the Quebrada Grande population, and subpopulations 2, 3 and Arroyo del Tanque of the Río Loco population in the Susúa Forest Reserve. In most cases, the plants for which GPS readings were not obtained were the last plants found. In the case of the Quebrada Peces population, readings were not obtained for any of the plants in tributaries C and F (Fig. 23). Also, a GPS reading could not be obtained for any of the plants in subpopulations 2 and 3 of the Río Loco population. The reading for these subpopulations was taken at the trail which provides access to the quebradas. In the case of the populations that extend from the forest reserve into private land, such as the Quebrada Peces and Quebrada Grande populations, there is not a clear idea of their precise location. This is an important concern because the species' endangered status does not protect individuals outside of forest reserve land.

The Río Bonelli (subpopulation 2) and Río Lajas populations, both in the Municipality of Maricao, were found on private lands, while the Quebrada Peces population, found in the Municipalities of Sabana Grande and Yauco, and the Quebrada Grande population in the Municipality of Yauco both extend from the Susúa Forest Reserve onto private lands. The sixteen individuals of the Río Bonelli subpopulation 2 and the nine individuals of the Río Lajas population were found outside the Maricao Forest Reserve, comprising 15% of the plants found

in Maricao. Fifty-five plants of the Quebrada Peces population (32% of the population) were found outside the Susúa Forest Reserve (tributaries A, D, E, Fig. 23). One of these plants was observed to have 18 fruits in August of 2007, the largest number we have ever observed. Nine individuals of the Quebrada Grande population (five in subpopulation 1 and four in subpopulation 2), 23% of the population, were found outside of the Susúa Forest Reserve. The area between subpopulations 1 and 2 of the Quebrada Grande population was not searched for individuals, which means that the percentage of plants found outside the reserve's boundary for this population and the forest may actually go down if more individuals were found. The area that was not searched is mostly on forest reserve land (Figs. 26 and 27). Seventeen percent of the plants in Susúa were found outside of the reserve's boundary, and 17% of all of the plants reported in this work were found growing outside both of the forest that were studied. The plants found outside the Maricao Forest Reserve were much farther from the forest boundary (up to ca. 792 m) than those found outside the Susúa Forest Reserve.

The New Law of Wildlife of Puerto Rico, Law Number 241 of August 15, 1999, specifies that its purpose is to protect, conserve and promote native and migratory wildlife species, and to declare as property of Puerto Rico all of the wildlife species in its jurisdiction. Article 3 of Law Number 241 states that the protection of these species and in particular its natural habitat is public policy of the Government of Puerto Rico. This law establishes the prohibition of the modification of habitats that are critical and essential to endangered and vulnerable species. Also, the DNER enforces the Law for the Conservation, the Development and the Use of Water Resources, Law Number 136 of June 3, 1976, which makes the margins of water bodies public domain and the DNER responsible for the protection of the margins of water bodies up to a distance of 5 m from the water body's legal channel (L. A. Vélez-Roché, pers. comm.). The legal

channel includes the channel or the natural canal and the shore. The shore includes the lateral strips of the canals of rivers which include the low and high water mark, and the margins as the lateral zones by the shore. This means that *Crescentia portoricensis* is protected under the New Law of Wildlife of Puerto Rico, and its habitat is protected by the Law of Water, because the species grows at the margins of water bodies.

Crescentia portoricensis is found in two river drainage basins in the Maricao Forest Reserve and in one drainage basin in the Susúa Forest Reserve. In the Maricao Forest Reserve, the Río Maricao, Quebrada Seca, Quebrada Piedras and Río Cupeves are in the Río Guanajibo drainage basin, while the Río Bonelli, Río Postrero, Río Lajas and Quebrada Negra are in the Río Grande de Añasco drainage basin. In the Susúa Forest Reserve, the Quebrada Peces, Quebrada Grande and the Río Loco are all in the Río Loco drainage basin. In the past, for fruits and seeds to have been dispersed to three river drainage basins that do not connect, a fruit and/or seed dispersal agent must have existed. Janzen (1982) proposed that the contents of the fruits of Crescentia alata in Costa Rica could have been dispersed by Equus fraternus, the Central American Pleistocene horse. On the other hand, Janzen cites Gentry's proposal that because of the fruits' fleshy-sweet interior they could have been dispersed by some unspecified mammal, but states that Gentry strongly believed that *Crescentia* is water dispersed. Fossil evidence has revealed that extinct mammals in Puerto Rico include one shrew, one sloth, three leaf-nosed bats and five rodents, which may have disappeared as a consequence of climatic changes in the Pleistocene Epoch, by their interaction with humans (modification of habitat and exploitation by both pre-Columbian people and Europeans) or by the interaction with rats, cats, dogs and mongooses (Gannon et al., 2005). For example, the leaf-nosed bats of the family Phyllostomidae

consume insects, leaves, fruit, pollen, nectar, blood, or small invertebrates, including birds, rodents, frogs, lizards, and other bats (Gannon et al., 2005).

#### **Changes in populations over time**

In 1996, Breckon and Kolterman reported 331 plants of *Crescentia portoricensis* in nine populations; this study reports 532 plants in 11 populations, an increase of 60% in the number of plants (Tables 2 and 3). At the time of their study, which lasted from 1991 to 1995, they were working with several different species. This research reports a larger number of plants because the search, specifically focused on *C. portoricensis*, was more extensive than the one done by Breckon and Kolterman. It is inferred that the previously unreported plants reported in this research may be old individuals, as no seedlings were found from 1991 to 1995 and no seedlings were found in the course of this research.

At the same time, no direct information is available on the growth rate of the plants. A comparison was made between the data recorded by Breckon and Kolterman and data recorded in this research for the Río Maricao population, Maricao Forest Reserve and the Arroyo del Tanque population, Susúa Forest Reserve. As is observed in tables 4 and 5, the growth rate of *Crescentia portoricensis* was found to be very low and also quite variable. Table 4 presents the diameters of five plants in the Río Maricao population, Maricao Forest Reserve in April of 1993 and in January of 2008, while Table 5 presents the diameters of 73 plants in the Quebrada Peces (26) and Quebrada Grande (2) populations and the Arroyo del Tanque subpopulation (45), Susúa Forest Reserve. The slower growth observed in the Maricao plants (Fig. 33) is unexpected, and may reflect the fact that comparative data were only available for five plants in the Río Maricao population in which the greatest loss of individuals was observed. On

the other hand, the mean numbers of stems from the base were similar in the two forests and also showed little change over time (Fig. 34), which suggests that stem production and stem loss occur at more or less the same rate. According to an ANOVA (Appendix V), there was not a significant difference (p=0.8014) between the mean growth per year in Maricao vs. Susúa.

Plants were considered to reproduce asexually when the stem and the branches rested on the ground, when the stem or branches became buried or when the tip of a branch came in contact with the ground. The percentage of individuals reproducing asexually in the Maricao and Susúa Forest Reserves appear in Table 7. The process of asexual reproduction was not observed in its final phase in any of the plants observed, because the stems and branches that had roots had not detached from the main plants to form new individuals. Plants should be monitored to see how long it takes them to grow roots at stems and branches that rest on the ground, and to see if the process of asexual reproduction is completed.

Tables 2 and 3 show that *Crescentia portoricensis* was found to exhibit few plants with reproductive structures (flowers, fruits, or both). Only 8.5% of the plants were observed in reproductive condition. A higher percentage of plants in Maricao (10%) than in Susúa (8%) were observed to be fertile; according to a G-test, this difference is not significant (p=0.463). In any case, the timing of visits to the different populations may affect the results. The percentage of individuals reproducing sexually in the Maricao and Susúa Forest Reserves are presented in Table 8. The Quebrada Piedras, Río Lajas and Quebrada Negra populations in the Maricao Forest Reserve were not found to include fertile plants, as well as the subpopulation 2 of the Quebrada Grande population, the Quebrada Calliandra subpopulation, subpopulation 2, and the Arroyo del Tanque subpopulation, all part of the Río Loco population, in the Susúa Forest Reserve.

Plant fertility may be affected by many biotic and abiotic factors. Factors such as pollinators, predators, soil fertility, temperature, precipitation, availability of sunlight, and competition for space with other plants, are crucial for plant fertility and seedling establishment. For example, Johnson and Sindel (2003) postulate that the reproductive strategy of a perennial weed depended strictly on the environment. It was found that the weed reproduced asexually when it was more probable that vegetative material established over seed, when there was heterogeneity in the environment in terms of unfavorable and favorable establishment sites and when there was a high number of establishment sites. When there was a high probability of seedlings establishing, when the available sites were homogeneously unfavorable and small, sexual reproduction was favored.

The primary reason for the loss of individuals is believed to be erosion of stream banks as a consequence of deforestation and poor management practices upstream, as observed by biologists of the Department of Natural and Environmental Resources working in the forest reserves (USFWS, 1991). Two populations previously known before 1979 in the Maricao Forest Reserve have disappeared due to flooding and erosion of their habitat (USFWS, 1991). Erosion may explain why the number of individuals in the Río Lajas (Maricao), Quebrada Seca (Maricao) and Arroyo del Tanque (Susúa) populations have decreased as compared to the data gathered by Breckon and Kolterman from 1991 to 1995.

The loss of 50 plants (60% of the population) in the Río Maricao population (Maricao) could be explained by the construction of a dam in the river. This dam was first constructed in 1938. Hurricanes destroyed the dam twice, the last time in 1998 (hurricane Georges). By 2000, reconstruction of the dam had been finished, which may have caused the water level to rise resulting in the loss of plants.

Another reason that could explain the loss of individuals is cutting. Four plants were found to be cut, two on private land, two on forest reserve land. A plant was found on private land in the Río Lajas population in Maricao, and another in the Quebrada Peces population just outside the Susúa Forest Reserve. The Quebrada Peces population extends from the forest reserve into private land. The other two plants were found in forest reserves, one in the population of Quebrada Negra in Maricao, which was apparently cut by employees of the DNER while reopening trails in the area, the other in the subpopulation of Arroyo del Tanque in the Susúa Forest Reserve. The Arroyo del Tanque plant (# 93) (Fig. 31), was cut while repairing the water tubing which collects water from Arroyo del Tanque. A subsequent visit was made nine months later to see if plant # 93 had resprouted, but it was dead. In September of 1991 Breckon and Kolterman found the trunk of plant #22 to be cut, but this plant was found to be alive in September of 2007.

### **Reproductive biology**

Asexual and sexual reproduction for plants of different size classes (based on stem diameter) are presented in Figures 35 and 36. The minimum stem diameter for asexual reproduction in the Maricao Forest Reserve was 1.2 cm (Fig. 35A), while for sexual reproduction it was 2.6 cm (Fig. 36A). These two plants were found in the R.ío Cupeyes population. In the Susúa Forest Reserve the minimum stem diameter for asexual reproduction was 1.0 cm (Fig. 35B) for a plant found in the Arroyo del Tanque subpopulation, while for sexual reproduction the minimum stem diameter was 1.3 cm (Fig. 36B) for a plant in the Quebrada Peces population. Although a few plants were found with smaller stem diameters than the smallest individuals observed with asexual or sexual reproduction, they probably do not represent juveniles or

immature plants. The sampling methods were not designed to identify all reproductive events, and almost surely did not do so. In addition, as mentioned elsewhere, seedlings of *Crescentia portoricensis* have never been observed in the wild.

Breckon and Kolterman (1994) conducted observations on phenological patterns in one or both forests during every month of the year between September of 1991 and August of 1994; but no reproductive structures were recorded for March and August. They observed that flower buds were most abundant in the months of February and January, and they reported the greatest number of flowers in the month of February and the second greatest number of flowers in the months of January and April. The highest numbers of fruits were observed in the months of October, February and April; of the three structures mentioned fruits were the most abundant. This is due to the fact that fruits last longer on the plants, while buds can be ripped off the plants by landslides, strong winds, strong water currents in events of heavy rain, or animals. Flowers that are not pollinated are discarded by the plant, or may even suffer the same fate as mentioned for the flower buds. In this work the majority of the flower buds were observed in January and March, while the majority of the flowers were observed in March and January. Fruiting was observed at its peaks in April and August, but in August in the Quebrada Peces population a plant was observed to have 18 fruits at the same time. The majority of the observations were recorded in January; in this month six populations were visited. No observations were recorded on the months of May and July.

As is recorded in Appendix IV, the flowers of *Crescentia portoricensis* last about nine days on the plant. The corolla was observed to take three days to open, but by that day the filaments and style were fully exserted and the anther and stigma were dull pink. By the fifth day the corolla had fallen, the style was still attached to the ovary, but the tip of the stigma appeared

black. On the seventh day the style had fallen from what was left of the flower and by day number nine the ovary was found on the ground. Fruits were observed to last approximately a year on the plant (Breckon and Kolterman, 1993).

Between August 1994 and August 1995, Breckon and Kolterman observed general phenological patterns of sporadic flowering that suggested that C. portoricensis probably flowers throughout most of the year (Breckon and Kolterman, 1996). This study suggests that C. *portoricensis* flowers throughout most of the year and that flowers last nine days on the plant. No flower visitors were observed, but the colors on the flower suggest that the flowers can be pollinated by bats. Schmitt (1983) suggests that changes in pollinator visitation may reflect weather, changes in pollinator population numbers, or a behavioral response to changes in flower density or the availability of competing flower species. At 7:08 AM, in the morning of December 24, 2007, the morning after the pollination observations were made, an unpleasant odor was smelled and a discharge of a transparent liquid was observed in the flower. It was assumed that this transparent liquid was a way to attract a pollinator and a reward that was not claimed by it. According to Tschapka and Dressler (2002) most bat-flowers are attracted to odors that are frequently perceived by humans as unpleasant and have been compared, for example, to overripe beans, sour milk, chlorine, mouse urine, human excrement, garlic or a cadaver. Agents of seed dispersal were not identified, even though termites were observed feeding on the remaining seeds of an old fruit that was found on the ground, but they were not observed carrying any of the seeds. Since no dispersers were observed it is assumed that seeds are dispersed by water.

### Hybridization

It is known that species of the genus *Crescentia* hybridize (Gentry, 1980), but no evidence of hybridization was found in any of the wild populations of *Crescentia portoricensis*, even though a plant of *C. alata* was found in front of the resident forest manager's residence in the Susúa Forest Reserve, right next to the Río Loco, and two plants of *C. cujete* were also found in that forest, one in the Río Loco, the other in the southern portion of Quebrada Peces. In Quebradillas, cultivated plants (see Appendix VI) of *C. portoricensis* and *C. linearifolia* were observed to have fruits of intermediate size and shape (M. A. Vives-Heyliger, pers. comm.), and at the Caguas Botanical Garden, five out of the seven *C. portoricensis* planted at the garden are apparently hybridizing, fruits appeared to look more like fruits of *C. cujete* than *C. portoricensis* (J. Golgiewicz, pers. comm.). Hybridization with other species of the genus should be avoided both *in situ* and *ex situ*, because *C. portoricensis* could be in danger of hybridizing out of existence. Hybridization can be determined by examining leaf and fruit characteristics based on Gentry's (1980) key [leaf size and shape, leaf surface (shiny vs. dull), fruit shape and size] and molecular studies.

#### **Genetic diversity**

The conservation of a rare species should include the preservation of its genetic diversity. To determine the extent and patterns of genetic diversity in *Crescentia portoricensis*, morphological and/or molecular methods should be used. Molecular studies could include iso/allozymes, DNA markers, or DNA sequences. An advantage of DNA sequences is that they could also be used to determine phylogenetic relationships among populations and species. A particular problem with rare species is the lack of genetic diversity (Falk, 1992; Fiedler and Ahouse, 1992; Rieseberg, 1991). This may be due to the founder effect or a genetic bottleneck during its history, such as may well have occurred as a result of massive deforestation in southwestern Puerto Rico prior to the establishment of the Maricao and Susúa forests. Another factor that would be expected to contribute to low genetic diversity in *C. portoricensis* would be its apparent reliance, at least at present, upon asexual reproduction.

In the management of a rare species, not only the overall level but also the patterns of genetic diversity should be taken into consideration. It is possible that all the genotypes present in the species as a whole are present in individual populations; this would be the case for an outcrossing species that reproduces sexually and exchanges genetic material through pollination and/or seed dispersal over its entire range. On the other hand, more limited gene exchange would be expected to result in genetic differences among populations. In the particular case of *C. portoricensis*, a preponderance of asexual reproduction would be expected to be reflected in genetic differences among river drainages. In addition, although the vegetation of Maricao and Susúa may well have been continuous in pre-Columbian times, subsequent fragmentation may have resulted in genetic difference between the two forests. Accordingly work is needed on the level of genetic diversity within and among forests, drainage basins and populations. If genetic diversity of the species is studied, we will have a clearer idea on how to reintroduce plants into existing and new populations.

#### Fruit and seed predation

In 1992 Breckon and Kolterman found two old fruits along a tributary of the Río Loco (Susúa) that had been chewed, presumably by the introduced rat (*Rattus rattus*). In this study a

fruit was found in Quebrada Peces (Susúa) to be chewed open, this fruit was green in color and still on the plant at the time of the visit. Janzen (1982) in a study done in Santa Rosa National Park in Costa Rica reported that the seeds of *Crescentia alata* were consumed by a small native terrestrial rodent (*Liomys salvini*); this rodent ate the seeds from horse dung. In laboratory experiments, *L. salvini* dug into a ball of the fruit's pulp, if it was given to it, and quickly ate the seeds, but could not open the hard fruits. Janzen thinks that the seeds might be protected from seed predators by the fruit's "hard fruit hull rather than toxins in the seeds." He also reported that on rare occasions, the variegated squirrel (*Sciurus variegatoides*) opened the ripe fallen fruits making a 3-5 cm diameter hole, scooped out the pulp and removed the seeds.

In 1993 Breckon and Kolterman reported that fruits in Quebrada Peces (Susúa) had been invaded by insects and appeared to contain no viable seeds. In this study, termites were observed feeding on the seeds from an old fallen fruit in Quebrada Peces. The lack of viability of seeds in the field could have been caused by predation; maybe the seeds do not have the opportunity to be dispersed outside the fruit because they are eaten while the fruit is still on the plant. If the fruits fall, the seeds that remain in the fruit can be quickly removed by insects and the ones that do fall out may suffer the same fate.

### **Conservation status**

The International Union for Conservation of Nature and Natural Resources Red List categories and criteria are "intended to be an easily and widely understood system for classifying species at high risk of global extinction" (IUCN, 2001). The system intends to offer an explicit and objective framework "for the classification of the broadest range of species according to their extinction risk" (IUCN, 2001). *Crescentia portoricensis* appears in the 1997 IUCN Red List

of Threatened Plants as <u>Endangered</u>, but the criteria are not included. It does not appear in the current online version of the Red List. Based on the present studies, the species includes 532 individuals, all of which are considered to be mature plants. The extent of occurrence is estimated at 43 km<sup>2</sup>, and the area of occupancy is estimated at 0.9 km<sup>2</sup>. At the very least, the species is <u>Vulnerable</u>: VU D1+2. It is important to monitor wild populations closely for any signs of decline, which could lead to its reclassification as <u>Endangered</u> under criteria B and C or <u>Critically Endangered</u> under IUCN criterion B.

According to the USFWS (1991), recovery plans "delineate reasonable actions which are believed to be required to recover and/or protect the species." The recovery plan of Crescentia portoricensis states that the actions needed to consider the downlisting of the species are: to monitor existing populations, provide protection for existing populations and their habitat, conduct research on the life history of the species, evaluate methods of propagation, and locate introduction sites, propagate and produce seedlings for enhancement of existing populations and for the establishment of new populations at identified sites. The plan for C. portoricensis needs to be reevaluated, and some actions need to be incorporated into the recovery plan based on the research done here. Genetic diversity should be studied, and genetically important populations should be protected to ensure diversity. Microhabitat requirements of the species should be assessed, as the survival of the species must be guaranteed. It is known that the species historically grows on serpentine and it is a moisture-limited shade-tolerant shrub (Breckon and Kolterman, 1994). It could be the case that if it were to be planted under other conditions, reintroduced plants might not survive or compete effectively with other plants. The only action needed to consider the downlisting of the species that has been partially met is the establishment of new populations and the enhancement of others. The Department of Natural and

Environmental Resources has planted 64 individuals in Quebrada Piedras, an unknown number of individuals in Quebrada Negra and between 75 to 120 plants in Finca Gabia and Toa Vaca, in the Municipality of Coamo, in recovery efforts for the species. Fruits were not observed in the Quebrada Piedras and Quebrada Negra populations throughout this research, but fruits were observed by DNER employees on the plants in Finca Gabia and Toa Vaca. The DNER and the USFWS did not evaluate microhabitat requirements or genetic diversity, and have not been monitoring the populations to check on the status of these populations. In addition, propagation has often been based on seeds from a single fruit, and record-keeping on the origin of plant material has been inadequate at best. It is clear that the USFWS criteria for downlisting, as presented in the Recovery Plan of *Crescentia portoricensis* have not been met, so the species should remain as Endangered. The increase in the number of populations and individuals presented in this research may not be a real increase but rather there may be a decrease. Breckon and Kolterman studied Crescentia portoricensis from 1991 to 1995, but they were working with several species at the time, so their search was not as extensive as the one done throughout this study. It is presumed that the unreported populations and individuals found throughout this study are mature, because the data recorded in this study suggests that C. portoricensis has a slow growth rate (Appendix V). In addition, seedlings have never been found in the wild.

#### **Management recommendations**

All of the populations need to be monitored and protected against human impacts. Plants on private lands should be protected against cutting, vandalism and over-collecting. The plants in the Río Lajas population, in particular, are in danger of disappearing, because they are in a heavily visited area (trail to Salto Curet). Four plants were found to be cut, and one of them was dead. Plants need to be protected against cutting, because it contributes to the loss of individuals or the reduction of their reproductive capability.

The DNER and the USFWS should seek to provide protection for the Río Bonelli population (subpopulation 2), Río Lajas population (Maricao), and for the plants that are outside of the Susúa Forest Reserve in the populations of Quebrada Peces and Quebrada Grande. The lands in which the plants are found can be acquired by the agencies and integrated into the existing reserves. Alternatively, the agencies could establish an agreement with the owners of the lands (conservation easement) in which the owners agree not to develop the lands and to fence off the areas in which the species occurs to stop any kind of cutting or vandalism.

These groups of plants should be categorized in terms of priority, based on the following criteria: genetic diversity and probability of survivorship. For example, if it is found that there is no significant difference in the genetic diversity among populations, then the agencies that are in charge of protecting the species must consider if it is worthwhile to acquire the lands in which the Rio Bonelli and the Rio Lajas populations are, and the lands in which the Quebrada Peces and the Quebrada Grande populations extend to, if the plants that are in these lands are not going to contribute any genetic variability. If these lands are acquired, if could mean an opportunity for the agencies to expand forest reserve land or possess lands with other rare or endangered species and ecological value. Also, these groups of plants could also be monitored to see if they are losing individuals. If these groups are losing individuals, then it would be up to the agencies to decide if populations that are outside the Maricao Forest Reserve, which account for 15% of the total plants reported for Maricao, are worth protecting, but this decision should be well thought out because there are only 163 plants reported for Maricao.

### Reintroduction

Plants can be introduced from seeds, cuttings, or transplant, but any reintroduction of the species should take into consideration genetic diversity and reintroduction sites. If needed, existing populations should be enhanced, but protected populations should be a priority. Genetic diversity should preferably be evaluated before doing propagations and reintroductions. Pending such information, reintroductions should be limited to the same forest, maybe the same river drainage or even the same population.

Experiments on seed germination have been done by Breckon and Kolterman at the University of Puerto Rico, Department of Biology's greenhouse; plants have also been grown from seed at the Fairchild Tropical Garden in Miami. According to Breckon and Kolterman (1994) the highest germination rates were observed under the conditions that prevail in the species' native habitat (wet to mesic soil under low to moderate light intensities). From the germination experiments and observations in the field, Breckon and Kolterman concluded that *Crescentia portoricensis* is shade-tolerant and moisture-limited. They also discovered that seeds from full-size green fruits germinate readily in the greenhouse.

Plants in Maricao were found along rivers and drainages on steep slopes which had an open and sparse canopy. The plants were almost completely exposed in the Maricao populations. Sites for reintroduction in Maricao should possess those characteristics. The Río Maricao site should be a priority for reintroduction due to the fact that the population lost 46 individuals, it is on the protected lands in the Maricao Forest Reserve, one must go through the DNER offices at the fish hatchery to access the population and it is the historical site for the species' discovery. Individuals should be planted in the Río Maricao population, taking into consideration that water levels rise due to heavy rain.

The Susúa plants were found to be along river banks and drainages under thick canopy and were not exposed. A promising site that should be evaluated for reintroduction in the Susúa Forest Reserve is the Quebrada Grande population, because it has the lower number of individuals compared to the other two populations in the forest reserve. Before making any reintroductions in Quebrada Grande, the area should be searched for plants between subpopulations 1 and 2. If Quebrada Grande is selected as a reintroduction site, planting should not be done in or near the private lands on which part of the population occurs.

Although *Crescentia portoricensis* grows in areas that differ widely in annual precipitation, its moisture tolerance may be rather specific. This species is found in more exposed areas, farther from rivers and streams, and on steeper slopes in Maricao than in Susúa. Local edaphic factors may also be important in providing an appropriate microhabitat for the plant.

At the time of reintroductions, one must take into consideration the type of soil. All of the populations reported in this work were found growing on serpentinite as the parental material and soils derived from it, but cultivated plants outside of the forest reserves are not necessarily growing on serpentine substrates. For example, in Quebradillas, cultivated plants are growing on Karst have even been observed to have fruits (M. A. Vives-Heyliger, pers. comm.). Also a healthy plant on the UPR-Mayagüez Campus is not on a serpentine-derived substrate. Evidently the species can survive on a variety of soils, but its competitive ability off serpentine substrates has not been evaluated.

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# CONCLUSIONS

- One hundred and 231 plants were reported for the Maricao and Susúa Forest Reserves respectively, as of 1996. This study reports 163 and 369 plants for the Maricao and Susúa Forest Reserves respectively. No seedlings were found in any of the wild populations of the species.
- A reduction was observed in the number of plants in the Río Maricao, Río Lajas, Quebrada Seca and Arroyo del Tanque populations, while numbers in the other populations increased.
- 3. Río Cupeyes and Quebrada Negra are two previously unreported populations found in the Maricao Forest Reserve. The Río Bonelli population in the Municipality of Maricao and the Quebrada Grande and Río Loco populations, located in the Susúa Forest Reserve, each have a previously unreported subpopulation.
- 4. Plants reproduce asexually when the stem and the branches rest on the ground and when the stem or branches get buried.
- 5. This study suggests that *Crescentia portoricensis* flowers throughout most of the year and that flowers last about nine days.
- 6. No flower visitors were observed, but characteristics such as the pale yellowish short bell-shaped corolla, the solitary flowers on lax branches and the nocturnal anthesis suggest that the flowers may be pollinated by bats.
- 7. No evidence of hybridization was found in any of the populations.
- Numbers of seeds ranged from 67 to 582 in fruits collected from different populations. Agents of fruit or seed dispersal were not identified, but it is inferred that fruits are primarily dispersed by water.

- 9. Fruits were observed to be chewed open even if they were not ripe, but agents of fruit predation were not observed. Termites were observed feeding on the seeds.
- 10. The Río Bonelli (subpopulation 2) and the Río Lajas populations, both in the Municipality of Maricao, were found in private lands, while the Quebrada Peces and the Quebrada Grande populations extend from the Susúa Forest Reserve into private lands.
- Plants were found to be cut in the forest reserve lands of Quebrada Negra and Arroyo del Tanque, and in the private lands of Río Lajas and Quebrada Peces.
- 12. The Department of Natural and Environmental Resources has planted 64 individuals in Quebrada Piedras, an unknown number of individuals in Quebrada Negra and between 75 and 120 plants in Finca Gabia and Toa Vaca in the Municipality of Coamo, in recovery efforts for the species.
- 13. This study suggests that *Crescentia portoricensis* is vulnerable to extinction, and its conservation status should continue to be monitored.

## RECOMMENDATIONS

- 1. New individuals and populations should be searched for in the Río Guaba, Río Bucarabones, Quebrada Culebra, Río Caín, Quebrada Agustina, and in all of the drainages between the intersection of Rd. 120 and Rd. 366 and the entrance of the DNER offices in the Maricao Forest Reserve, all in the Municipality of Maricao, Río Grande in the Municipality of Sabana Grande, Río Coco, Río Cañas, Quebrada Fría, Quebrada Mango Prieto and between subpopulation 1 and subpopulation 2 of the Quebrada Grande population, all in the Municipality of Yauco.
- 2. Further studies should be conducted of the reproductive phenology (periodicity, frequency, timing, and duration of flowering and fruiting).
- 3. Information on pollination should be gathered. Observations of flower visitors and studies on breeding systems should be conducted. Sampling should not be conducted only during short periods of time, as was done in this study, because the most likely pollinators (i.e., bats) are known to visit flowers sporadically and for short periods of time.
- 4. A study of the duration and development of fruits is of great importance so that mature fruits can be collected before they are predated. Data should be recorded on fruit and seed dispersal (including possible secondary dispersal). Fruits could be marked and observed to see if they are water-dispersed. Fruits should be observed for the identification of agents of fruit and seed predation.
- 5. Work is needed on the level of genetic diversity within and among populations. Both morphological and molecular data could be recorded, genetically important populations should be protected to ensure diversity.

- 6. The species should be propagated by seeds or cuttings. The Department of Natural and Environmental Resources, the U. S. Fish and Wildlife Service and volunteers of the agencies, universities and adjacent communities should work together in the propagation and reintroduction of the species.
- 7. *Crescentia portoricensis* should be reintroduced in its natural habitat in protected lands. Known populations could be enhanced if needed, taking into consideration the plant's genetic diversity. The species should be introduced in places where *C. cujete* and *C. alata* are absent. Hybridization with any other species of the genus should be avoided.
- 8. Existing populations should be monitored within and outside protected lands, especially in heavily visited places. Plants should be taken into consideration when repairs are made to water tubing, the opening of trails or any kind of development.
- 9. The public (schools near the forest reserves and forest visitors) and the employees that work for the Maricao and Susúa Forest Reserves and the "Compañía de Parques Nacionales" should be educated. These employees should be taught to recognize at least listed plant species to prevent cutting.
- 10. Any heavily visited areas in which the species occurs should be fenced off to minimize any cutting, over-collecting or vandalism.
- 11. The private lands in which the species occurs (36% of populations and 17% of individuals) should be acquired and protected by the DNER and the USFWS. An agreement could also be made between both agencies and the owners of the lands in order to protect the plants.
- 12. The microhabitat requirements of the species should be assessed, as the survival of the species must be guaranteed.

13. To ensure the long-term survival of the species, efforts should be made to promote seedling regeneration in natural populations.

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# TABLES

**Table 1.** Historical collections of *Crescentia portoricensis* according to the BRAMHS database at MAPR. Historical collections are defined as those collected more than 35 years ago. Herbarium abbreviations follow *Index Herbariorum*.

Collector(s), Number	Date	Locality, Habitat, etc.
Britton, Stevens and Hess # 2455 ( <u>TYPE</u> ) – NY	April 2, 1913	Río Maricao, between 500 and 600 m in elevation.
Britton, Cowell and Brown # 4523 – NY	February 15, 1915	Indiera Fría, Maricao, along river. Vine- like shrub, at an elevation of 430 to 800 m.
L. R. Holdridge # 460 – NY	June 6, 1936	Maricao Forest Reserve. Small tree.
L. R. Holdridge # 738 – UPR	May 28, 1941	Susúa Forest Reserve, along Río Loco, above Susúa camp, north of Yauco. 2.4 m shrub with a fruit.
Cobin # 1210 – MAPR	May 23, 1944	Maricao station grounds, wooded area.
A. González-Más # 1717 – MAPR	July, 1961	Maricao Forest Reserve.
R. O. Woodbury s.n. – NY	May 28, 1963	Monte del Estado. Maricao Forest Reserve.
R. O. Woodbury s.n. – NY	July, 1970	Maricao.

			er of Main St om the Base	tems	Main Sten	n Diamete	er (cm)		
Population	N	range	mean	s.d.	range	mean	s.d.	% Fertile	Elevation (m)
Río Maricao	38	1-6	1.8	1.13	0.8-4.0	2.30	0.88	10	490-575
Quebrada Seca	5	1-3	1.4	0.89	1.6-5.7	3.32	1.30	40	707-716
Quebrada Piedras	9	1-2	1.4	0.53	0.9-3.0	1.91	0.86	0	804-834
Río Bonelli	16	1-4	1.8	1.11	0.7-4.7	2.49	1.12	25	273-448
Río Cupeyes	27	1-3	1.3	0.59	0.6-5.2	1.77	0.84	15	678-690
Río Postrero	2	2-4	3.0	1.41	1.9-4.8	3.19	0.98	100	730
Río Lajas	9	1-3	1.3	0.71	0.7-3.5	1.87	0.87	0	414
Quebrada Negra*	57	1-7	1.8	1.13	0.6-7.4	3.05	1.02	3.5	814-817
Total	163	1-7	1.7	1.02	0.6-7.4	2.51	1.09	11	273-834

Table 2. Population structure and size distribution for Crescentia portoricensis Britton in the Maricao Forest Reserve.

\*According to Mr. Padrón-Vélez the population is a combination of wild plants and an unknown number of planted individuals.

		Number of Main Stems from the Base			Main Stem Diameter (cm)				
Population	N	range	mean	s.d.	range	mean	s.d.	% Fertile	Elevation (m)
Quebrada Peces									
Quebrada Peces North	113	1-8+	1.4	1.03	0.9-7.6	2.81	1.22	9.7	158-292
Quebrada Peces South	57	1-4	1.3	0.66	0.8-7.3	2.44	1.43	21	159-237
Quebrada Grande									
Subpopulation 1	35	1-3	1.8	0.84	0.8-6.8	2.44	1.14	2.9	134-305
Subpopulation 2	4	1-3	1.8	0.96	1.2-3.6	2.25	1.55	0	227
Río Loco									
Quebrada Calliandra	14	1-3	1.4	0.63	1.0-7.4	2.74	1.58	0	136-222
Subpopulation 2	41	1-4	1.4	0.62	0.9-7.7	2.43	1.34	0	281
Subpopulation 3	19	1-5	1.8	1.23	0.7-4.6	2.48	1.01	10.5	240
Arroyo del Tanque	71	1-5	1.8	0.98	0.3-5.4	2.19	1.08	0	137-234
Subpopulation 5	15	1-3	1.2	0.58	1.8-5.9	3.38	1.03	33	127-145
Total	369	1-8+	1.5	0.91	0.3-7.7	2.53	1.24	8	127-305

**Table 3.** Population structure and size distribution for *Crescentia portoricensis* Britton in the Susúa Forest Reserve.

	March 4, 1	1993	January 11, 2008				
Tag number	Number of Main	Main Stem	Number of Main	Main Stem			
	Stems from the Base	Diameter (cm)	Stems from the Base	Diameter (cm)			
318	2	1.4, 3.0	2	2.9, 3.2			
325	2	3.4, 4.0	1	4.0			
328	2	2.5, 2.5	1	3.0			
339	1	2.0	1	2.5			
341	1	5.5	2	3.37, 3.41			

**Table 4.** Number of main stems from the base and main stem diameter on April 3, 1993 and on January 11, 2008 for previously tagged individuals in the Río Maricao population, Maricao Forest Reserve.

	Number of Main	Main Stem	Number of Main	Main Stem
Tag number	Stems from the Base	Diameter (cm)	Stems from the Base	Diameter (cm)
uebrada Peces				
	October 19	, 1991	June 2'	7, 2007
109	1	2.2	1	2.32
114	1	2	2	2.89, 2.05
115	1	1	1	2.71
123	1	1.5	1	1.94
124	1	1.5	1	1.96
125	1	1	1	2.19
128	1	2.5	1	2.95
129	1	1	1	1.51
133	1	2	1	3.94
134	1	1.5	1	3.19
135	1	2	1	2.67
137	1	1	1	1.18
138	1	1.5	1	2.12
139	1	2	1	2.21
140	1	1.3	1	2.15
142	1	2	1	2.35
143	1	1.5	1	1.6
146	1	1.3	1	1.6
147	1	1	1	1.21
148	1	2	1	1.85
149	1	2	1	2.35
150	1	2	1	1.66

**Table 5.** Number of main stems from the base and main stem diameter for previously tagged individuals in the Quebrada Peces and Quebrada Grande populations and in the Arroyo del Tanque subpopulation, Susúa Forest Reserve.

152	1	3.2	2	3.23, 3
154	1	3.2	2	3.72, 2.21
156	1	4	1	4.07
157	1	2.5	1	2.57
Quebrada Grande p	opulation			
	Februa	ary 1, 1992	Ja	nuary 7, 2008
204	1	2.9	1	3.54
216	1	4.95	3	2.37, 2.51, 4.25
Río Loco populatio	n			
Arroyo del Tanque				
	Septem	ber 14, 1991	Ser	otember 7, 2007
4	1	2	1	2.32
6	1	2	3	2.3, 2.2, 1.8
8	4	2.5, 2, 1.5, 1.5	4	3.2, 1.88, 1.71, 1.84
9	1	1.5	1	1.65
10	1	1	1	0.91
11	1	1.5	1	1.06
12	3	2.5, 2, 1	2	2.04, 3.04
14	1	2	1	1.8
15	1	1.2	3	1.54, 2.59, 1.7
18	1	2.2	2	0.5, 2.33
21	1	1	1	1.26
22	1	4	2	4.03, 1.51
23	2	2.2, 1.5	2	1.74, 3.3
25	1	2.5	2	2.91, 2
26	1	2.5	2	2.24, 1.48
27	1	1.5	1	1.8

28	1	1.7	1	1.8
29	1	3	1	1.8
35	1	2.5	1	2.43
36	1	1.2	2	1.46, 1.94
38	1	2	2	2.76, 2.43
39	1	2.5	2	3.1, 2.93
40	1	2.2	2	0.93, 1.26
51	1	1.2	3	2.44, 2.16, 6.02
42	1	1.5	1	1.57
43	1	3.5	1	3.07
44	1	2	2	3.95, 2.06
45	1	2.5	3	3.4, 2.71, 2.23
46	1	4	2	3.52, 4.61
48	1	3.5	5	3.55, 3.51, 4.6, 0.66, 0.91
50	1	1.5	1	2.2
54	1	1.5	1	1.32
55	1	1	4	1.17, 1.86, 1.32, 0.92
56	1	2	2	0.72, 1.81
57	1	1.2	2	0.92, 1.46
58	1	1.2	1	1.64
63	1	2.5	1	3.27
64	1	2	1	4
65	1	2.5	1	3.03
67	1	1	2	1.18, 0.73
68	1	2.8	1	2.34
70	1	2	4	2.0, 2.76, 2.04, 1.09
71	1	2.5	3	3.04, 2.1, 1.73
91	1	2.7	2	2.98, 3.14
96	1	2.8	1	2.98

		Mean Frui	t Lengtł	n (cm)	Mean Fru	it Diamet	er (cm)	Length:	Width R	atio
Population	Ν	range	mean	s.d.	range	mean	s.d.	range	mean	s.d.
MARICAO										
Río Maricao	1	9	9	-	2.8	2.83	-	3.19	3.19	-
Río Bonelli	2	12.0-12.2	12.11	0.16	3.3-3.4	3.36	0.04	3.60-3.62	3.61	0.01
Río Cupeyes	3	7.8-8.7	8.31	0.45	3.6-4.2	5.09	0.32	2.02-2.20	2.13	0.1
SUSÚA										
Quebrada Peces North	15	5.0-13.4	9.36	2.38	1.3-5.8	3.14	0.95	1.25-4.10	3.15	0.87
Quebrada Peces South	16	7.2-12.9	10.07	1.63	2.3-7.2	3.79	1.67	1.61-4.14	3.00	0.92
Quebrada Grande Subpopulation 1	2	10.5-12.0	11.23	1.03	3.1	3.11	0.01	3.37-3.85	3.61	0.34
Río Loco Subpopulation 3	3	9.4-10.4	9.80	0.53	2.8-3.8	3.17	0.58	2.44-3.55	3.17	0.63
Río Loco Subpopulation 5	3	8.0-10.6	8.75	1.60	2.1-3.1	2.62	0.52	2.87-3.86	3.38	0.5

**Table 6.** Number of fruiting plants and mean size of fruits per plant for populations of *Crescentia portoricensis* Britton. N = number of fruits. All fruits were  $\geq$  5 cm long.

	Total number	Percentage of individuals
Population	of individuals	reproducing asexually
Maricao Forest Reserve		
Río Maricao	38	11
Quebrada Seca	5	20
Río Bonelli	16	25
Río Cupeyes	27	11
Río Postrero	2	50
Quebrada Negra	57	3.5
Overall	145	9
Susúa Forest Reserve		
Quebrada Peces	170	9
Quebrada Grande	39	10
Río Loco	160	12
Overall	369	10

**Table 7.** Percentage of individuals reproducing asexually in the Maricao and Susúa Forest Reserves.

\*Fifty percent of the plants in the Río Postrero and 2% of the plants in Quebrada Peces were reproducing both asexually and sexually. According to a G-test, there was not a significant difference (p=0.697) between the frequencies of asexual reproduction in Maricao vs. Susúa.

	Total number	Percentage of individuals
Population	of individuals	reproducing sexually
Maricao Forest Reserve		
Río Maricao	38	10
Quebrada Seca	5	40
Río Bonelli	16	25
Río Cupeyes	27	15
Río Postrero	2	100*
Overall	88	10
Susúa Forest Reserve		
Quebrada Peces	170	13
Quebrada Grande		
Subpopulation 1	35	3
Río Loco		
Subpopulation 3	19	10
Subpopulation 5	15	33
Overall	239	8

**Table 8.** Percentage of individuals reproducing sexually in the Maricao and Susúa Forest Reserves. Sexual reproduction refers to the presence of buds, flowers and/or fruits on the date on which each population was visited.

\*This population consisted of only two plants. A higher percentage of plants in Maricao (10%) than in Susúa (8%) were observed to be fertile; according to a G-test, this difference is not significant (p=0.463). In any case, the timing of visits to the different populations may affect the results.

# **FIGURES**



Figure 1. Crescentia portoricensis Britton [Bignoniaceae].



Figure 2. Yellowish-green flower of *Crescentia portoricensis*.



Figure 3. Bell-shaped flower of *Crescentia portoricensis*.



**Figure 4.** The yellowish-green bell-shaped flowers of *Crescentia portoricensis* ripen into dark green fruits.



**Figure 5.** The leathery leaves appear mostly in fascicles of two or three in *Crescentia portoricensis*.



Figure 6. Maricao Forest Reserve (M) and Susúa Forest Reserve (S).



Figure 7. The Maricao Forest Reserve (far back).



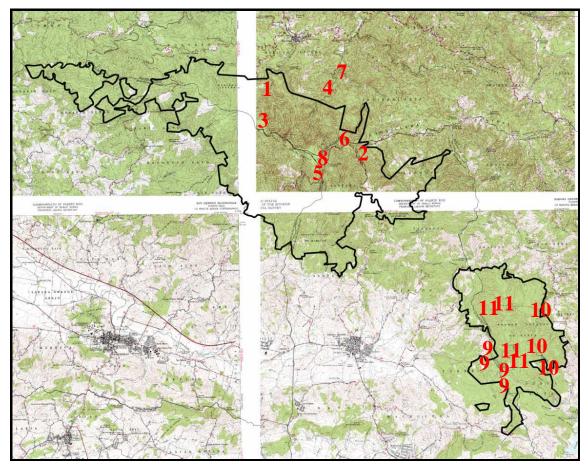
Figure 8. The Susúa Forest Reserve.



Figure 9. Young flower bud of *Crescentia portoricensis*.



Figure 10. Flower bud of *Crescentia portoricensis* near anthesis.



**Figure 11.** Populations are numbered from 1 to 8 for the Maricao Forest Reserve and from 9 to11 for the Susúa Forest Reserve.

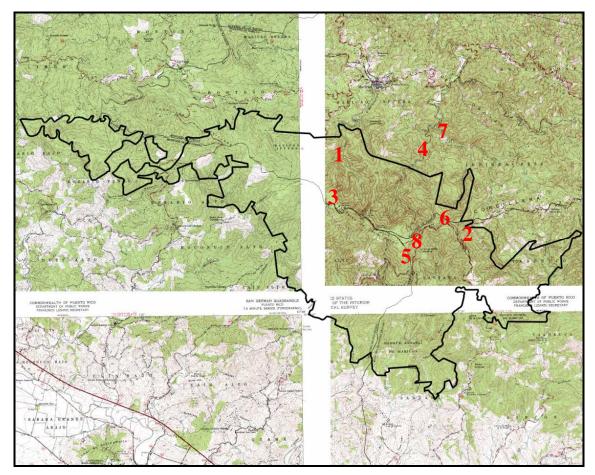


Figure 12. Populations are numbered from 1 to 8 for the Maricao Forest Reserve.

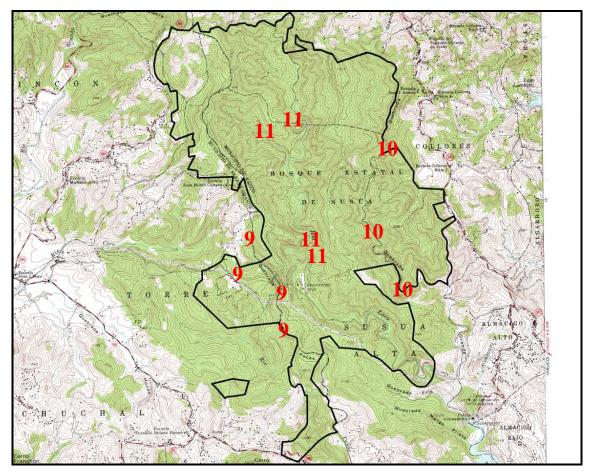


Figure 13. Populations are numbered 9 to11 for the Susúa Forest Reserve.

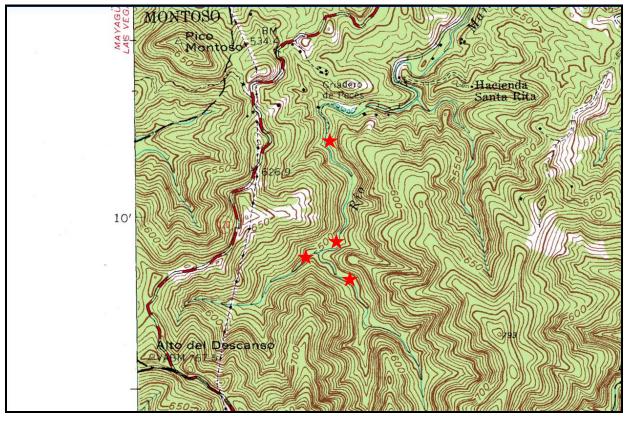


Figure 14. Río Maricao population, Maricao Forest Reserve. Stars mark the limits of the population.

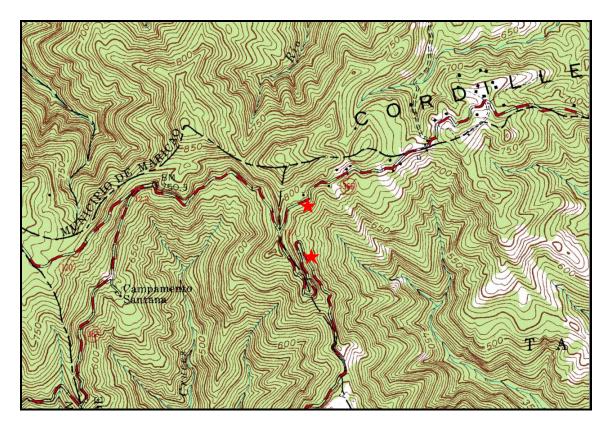
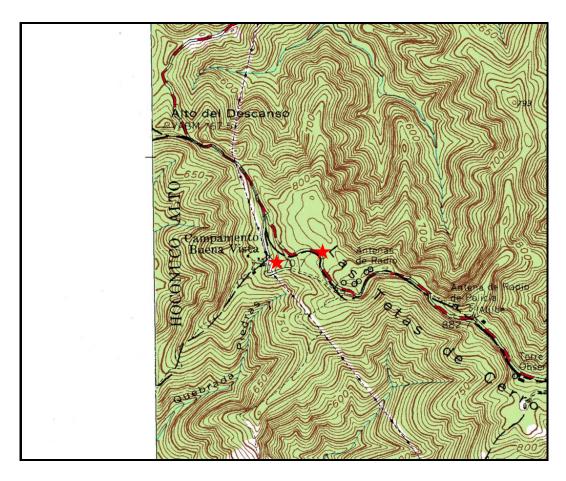
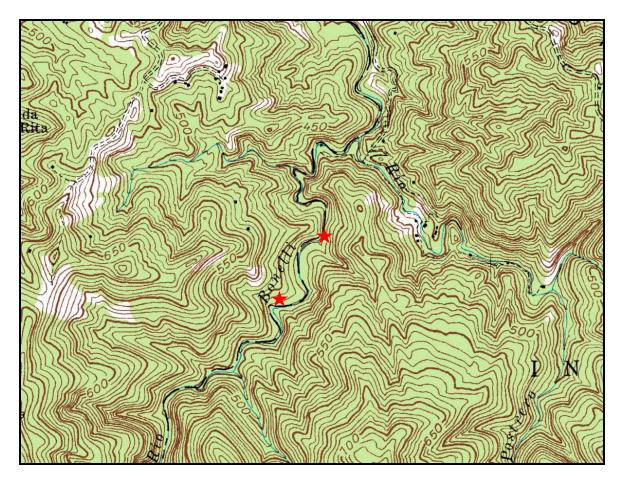


Figure 15. Quebrada Seca population, Maricao Forest Reserve. Stars mark the limits of the population.



**Figure 16.** Quebrada Piedras population, Maricao Forest Reserve. Stars mark the limits of the population.



**Figure 17.** Río Bonelli population, Maricao Forest Reserve. Stars mark the limits of the population.

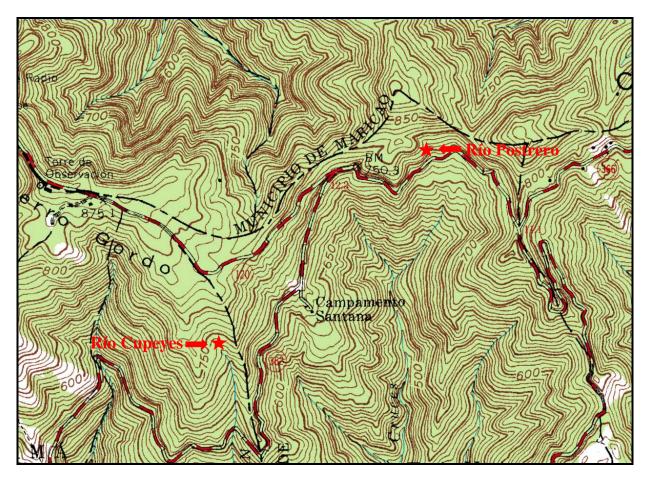


Figure 18. Río Cupeyes and Río Postrero populations, Maricao Forest Reserve.

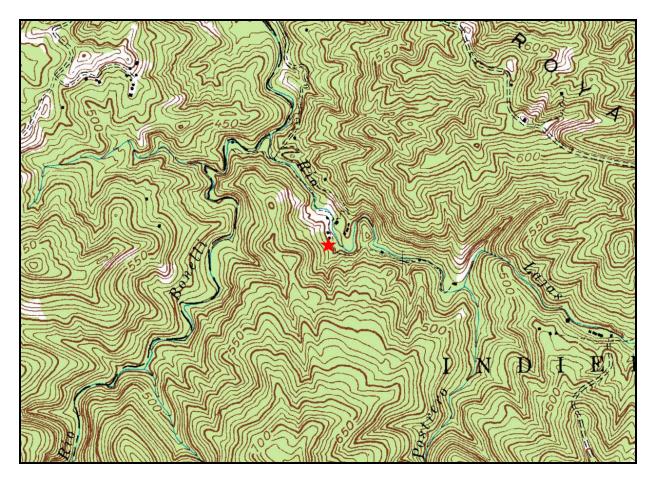


Figure 19. Río Lajas population, Maricao Forest Reserve.



**Figure 20.** PVC tubing that collects water from the Río Lajas, shown in the photo above. A plant of *Crescentia portoricensis* was cut to make space for the tubing. A plant of *Crescentia portoricensis* can be seen at the top left of the photo, very close to the tubing.

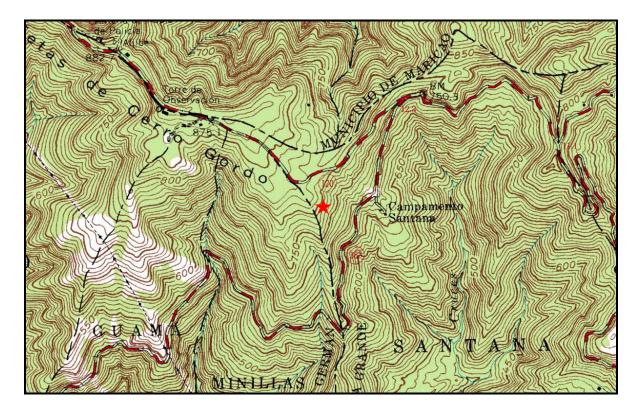
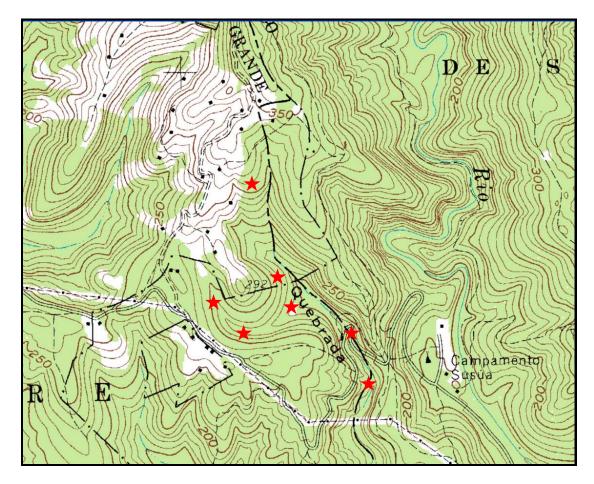
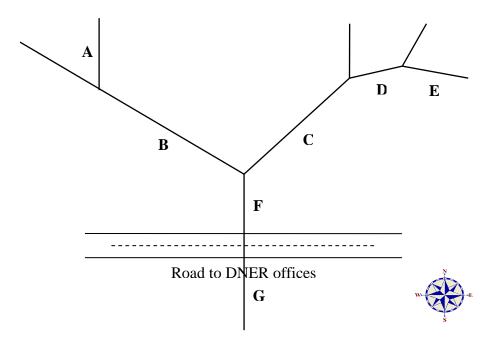


Figure 21. Quebrada Negra population, Maricao Forest Reserve.



**Figure 22.** Quebrada Peces population, Susúa Forest Reserve. Stars mark the limits of the population.



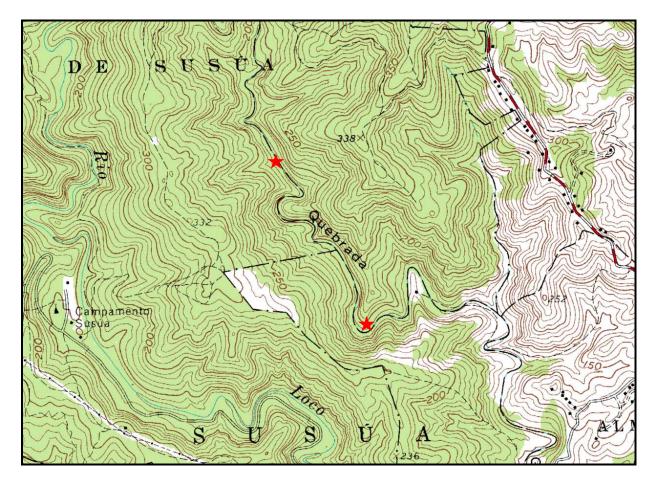
**Figure 23**. Number of plants by tributary (represented by letters) in Quebrada Peces, Susúa Forest Reserve. A = 19 individuals, B = 9 individuals, C = 33 individuals, D = 32 individuals, E = 11 individuals, F = 9 individuals and G = 57 individuals.



**Figure 24.** Three dark green fruits grow on the same branch from a plant in the south group of the Quebrada Peces population.



**Figure 25**. On a slope behind plant #117, an untagged plant had a small branch 0.64 cm in diameter connected to two stems. The branch had not detached to form a separate individual.



**Figure 26.** Quebrada Grande subpopulation 1 population, Susúa Forest Reserve. The red star in the south is where the first individual in the subpopulation was observed in the quebrada, the red star in the north represents the last GPS coordinates that were observed, but additional plants were found beyond this point.

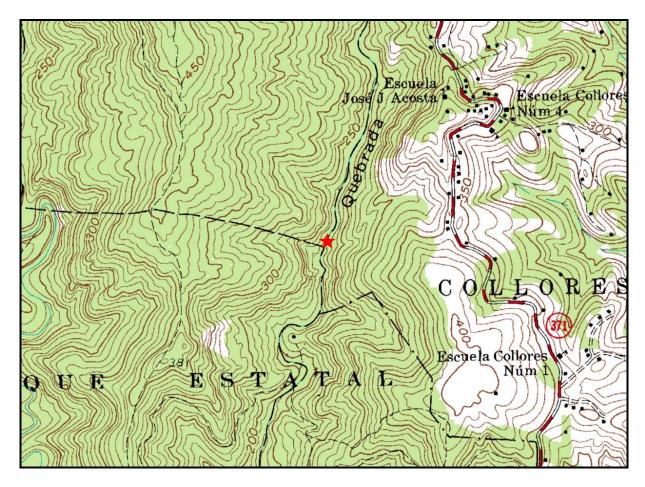


Figure 27. Quebrada Grande subpopulation 2 population, Susúa Forest Reserve.

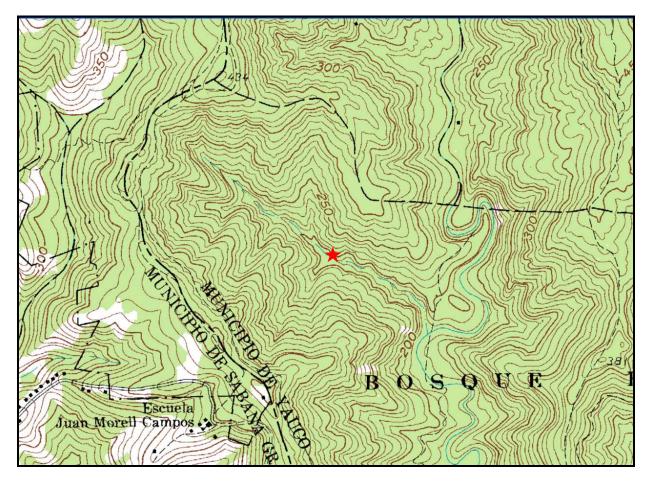


Figure 28. Quebrada Calliandra subpopulation, Susúa Forest Reserve.

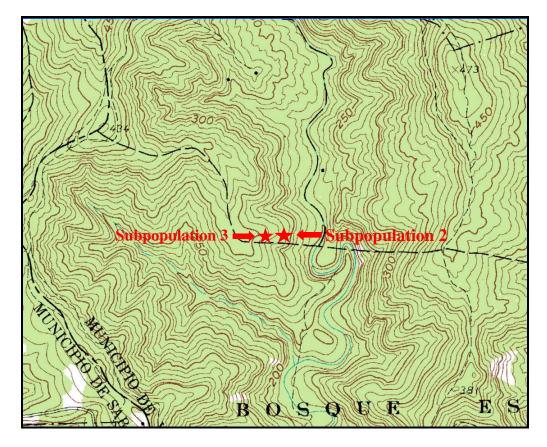


Figure 29. Río Loco 2 and 3 subpopulations, Susúa Forest Reserve.

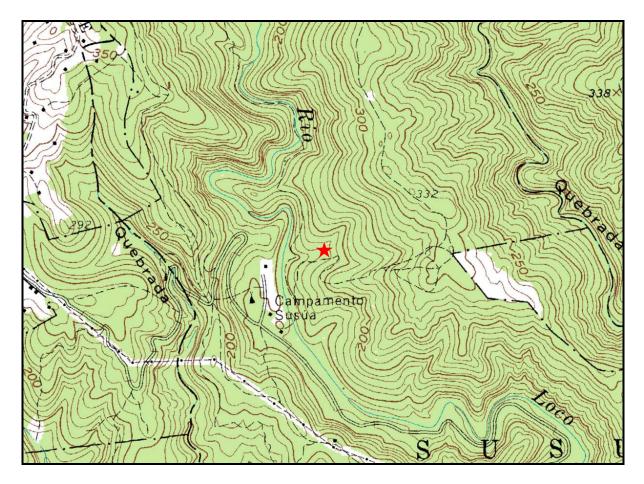


Figure 30. Arroyo del Tanque subpopulation, Susúa Forest Reserve.



Figure 31. Plant #93 in the Arroyo del Tanque subpopulation, Susúa Forest Reserve.

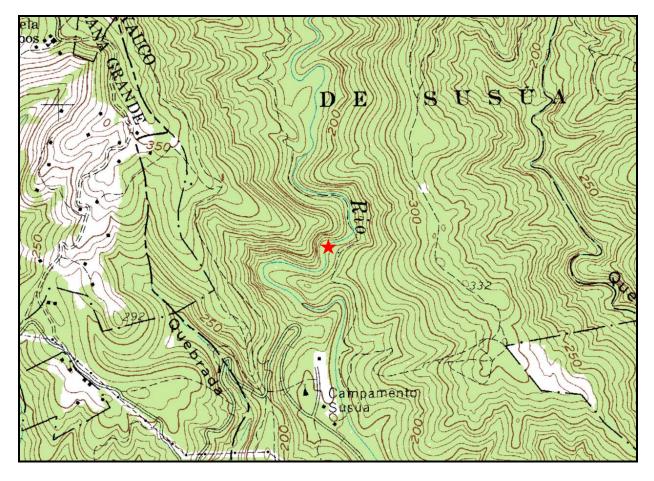
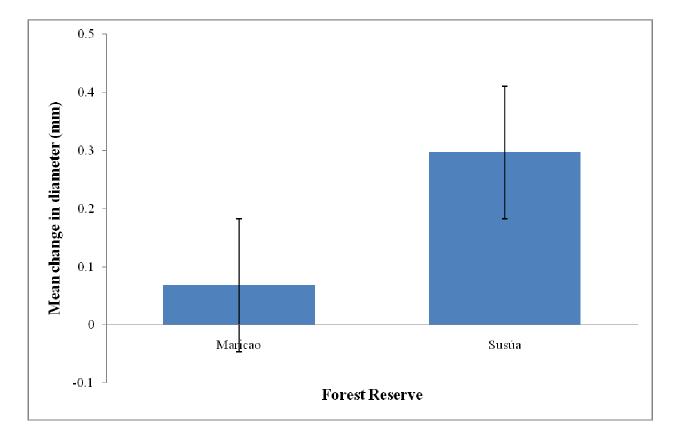
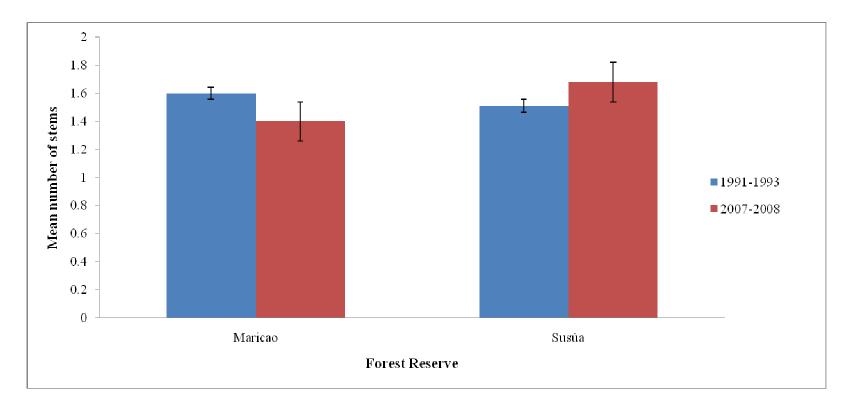


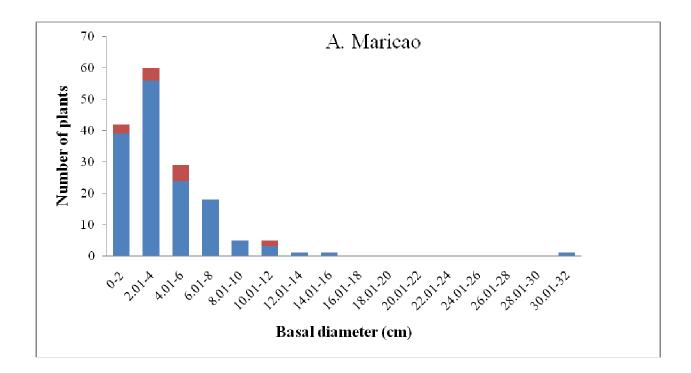
Figure 32. Río Loco Group 5 subpopulation, Susúa Forest Reserve.

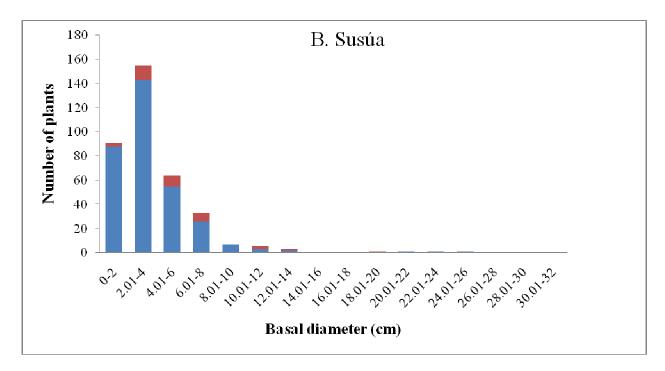


**Figure 33.** Mean change in diameter in the Maricao and Susúa Forest Reserves. Only the Río Maricao population is represented for the Maricao Forest Reserve, and only the Quebrada Peces and Quebrada Grande populations and the Arroyo del Tanque subpopulation are represented for the Susúa Forest Reserve.

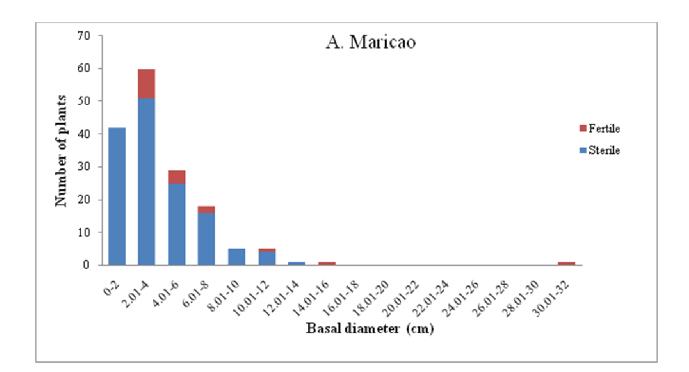


**Figure 34.** Mean number of stems in *Crescentia portoricensis* in the Maricao and Susúa Forest Reserves. Only the Río Maricao population is represented for the Maricao Forest Reserve, and only the Quebrada Peces and Quebrada Grande populations and the Arroyo del Tanque subpopulation are represented for the Susúa Forest Reserve.





**Figure 35.** Asexual reproduction in *Crescentia portoricensis*. Red bars represent individuals reproducing asexually, while blue bars represent individuals that are not reproducing asexually.



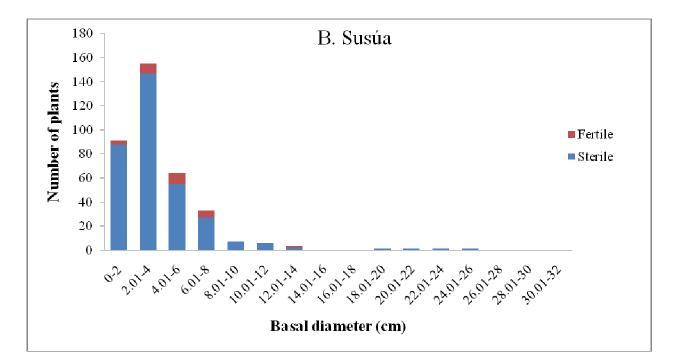


Figure 36. Sexual reproduction in Crescentia portoricensis.

# **APPENDICES**

Appendix I. Abbreviations.

Abbreviation	Significance
Agencies	
USGS	United States Geological Survey
DNR	Department of Natural Resources
USFWS	United States Fish and Wildlife Service
DNER	Department of Natural and Environmental Resources
USDA	United States Department of Agriculture
Herbaria	
MAPR	University of Puerto Rico, Mayagüez Campus
NY	New York Botanical Garden
UPR	Botanical Garden of the University of Puerto Rico
Others	
BRAHMS	Botanical Research and Herbarium Management System
Bo.	Barrio

Appendix II. Personal communications.

Person	Occupation
A. Muñiz-Suárez	Resident forest manager at the
	Department of Natural and
	Environmental Resources, Maricao
	Forest Reserve.
I. Sastre De Jesús	Professor at the Biology Department, University of Puerto Rico-Mayagüez Campus.
W. Cordero	Resident forest manager at the
	Department of Natural and
	Environmental Resources, Susúa Forest Reserve.
M. Justiniano	Environmental Educator at the
	Department of Natural and
	Environmental Resources-Mayagüez
	Regional Office.
G. J. Breckon	Professor at the Biology Department,
	University of Puerto Rico-Mayagüez Campus.
M. Rivera	Biologist at the Department of the
	Interior, U.S. Fish and Wildlife
	Service, Caribbean Field Office.
A. Rodríguez-Durán	Dean for Research, Inter-American
	University-Bayamón Campus.
L. A. Vélez-Roché	Director of the Administración de
	Reglamentos y Permisos (ARPE),
	2000-2008.
M. A. Vives-Heyliger	Ad Honorem Investigator of Puerto
	Rican Flora, University of Puerto
	Rico.
J. Golgiewicz	Arborist at the Caguas Botanical
	Garden.

**Appendix III.** Information on fruit (color and size) and seeds per fruit gathered for ten fruits, two from one plant from the Río Bonelli, Maricao Forest Reserve and eight from three plants from the Quebrada Peces, Susúa Forest Reserve.

Locality	Plant number	Color,	Number
		fruit size (cm)	of seeds
Río Bonelli, Maricao. 14.4 m upstream from east side of the river.	18° 09' 57.9" N, 066° 58'	04.7" W. Plant was grow	wing in a valley on the
	untagged	Dark green, 12.2 x 3.4 Dark green, 12.0 x 3.3	276, of which 20 seemed smaller. 267, of which 29 seemed smaller.
Quebrada Peces, Susúa No GPS data, but see l			
	124	Green, 10.0 x 3.3	275
Quebrada Peces, Susúa 18° 04' 3.3" N, 066° 5	a, south population. '4' 35.1" W, elev. 159 m		
	untagged	Black, 11.3 x 3.1	287
Quebrada Peces, Susú: 18° 04' 26.0" N, 066° :	a, north population. 54' 50.4" W, elev. 217 m		
	untagged	Dark green, 12.5 x 3.3	487
		Dark green, 12.3 x 3.3	217
		Dark green, 13.4 x 3.6	582
		Dark green, 10.2 x 2.5	104
		Dark green, 10.2 x 2.5	67

Quebrada Peces, Susúa, south population. 18° 04' 16.6" N, 066° 54' 36.7" W, elev. 178 m

untagged	Green, 12.9 x 3.9	362

All of the seeds were given to the DNER forest reserve managers. Seeds from Quebrada Peces were planted on August 31, 2007 in an alluvium, domesticated rabbit feces and sawdust medium. According to Wetsy I. Cordero-Nazario almost all of the seeds germinated; a thousand plants were given to the DNER's nursery located in the Cambalache Forest Reserve, Arecibo. One hundred-twenty plants remained in Susúa, these were going to be planted in September of 2009. Fruits collected in the Río Bonelli population were taken from the same plant, and all five fruits collected in the second location of the Quebrada Peces population shown in the table above were taken from the same plant.

**Appendix IV.** Observations made daily on flower longevity. These were done for 13 days (from 18 to 30 of December, 2007; mostly from 8:30 to 9:30 AM) in the subpopulation 5 of the Río Loco population. Observations from 1 to 9 correspond to the first flower, observations from 4 to 13 correspond to the second flower.

- 1. Corolla was not fully open, but it was bright green.
- 2. Filaments and style not fully exserted, corolla was bright green with white tips.
- 3. Filaments and style fully exserted, anther and stigma were dull pink, corolla was bright green with white tips.
- The anther and the stigma were black at its margins, the tips of the flower were yellow.
   Second bud started to open, the corolla was bright green.
- 5. Corolla from the first flower fell, style was still on the ovary. The tip the stigma appeared black. Ovary looked green in color, while the ring around the ovary was bright yellow. On the second flower observed, the filaments were not fully exserted, the style was fully exserted from the bottom of the corolla, the corolla was bright green with white tips.
- 6. Observations on flower visitors were made on the night of December 23, 2007; no visitors were observed. The anthers were starting to look black, flower corolla appeared yellow at its tips to 1.3 cm to the inside of the corolla, the rest of the corolla was green. At 7:08 AM the flower discharged some kind of substance which began to smell unpleasant. The stigma of the first flower seemed completely black.
- 7. The style fell from the first flower, ovary looked green in color. The calyx from that flower had become black at its margins. In the second flower the stigma and the anthers had become black at its margins. The corolla was yellowish green.
- 8. The tip of the ovary of the first flower was starting to turn black. Its ovary was still green in color while the ring around it was yellow. On the second flower, the corolla had fallen;

the stigma was starting to turn black. The ovary was green and the ring around it yellow, ants were observed inside of what was left of the flower.

- 9. The ovary of the first flower was found on the ground. The tip of the second flower's stigma appeared black, while the ovary looked light green, and the ring around it yellow.
- 10. The stigma of the second flower appeared black, while its style looked yellow. The ovary looked green, and the margins of the calyx looked black.
- 11. The stigma of the second flower appeared darker, while its style seemed to have curved upward a bit. The ovary looked green, and the margins of the calyx looked darker.
- 12. The stigma of the second flower was on the plant, it was black and looked if it might have moved to the right. The ovary looked green and the ring around it yellow. The calyx was darker at its margins.
- 13. The calyx of the second flower fell.

Appendix V. ANOVA of the mean growth per year in Maricao vs. Susúa.

New table: 11/19/2009 - 11:19:30 AM

Analysis of variance				
Variable	N	R 2	Adj. R²	CV
Growth per year (mm)	107	0.02	0.00	203.76

### (Type III SS)

F.V.	SS	df	MS	F	p-value
Model	0.56	4	0.14	0.41	0.8014
Location	0.56	4	0.14	0.41	0.8014
Error	34.61	102	0.34		
Total	35.16	106			

#### Contrasts

Location	SS	df	MS	F	p-value
Mar vs. Sus	0.06	1	0.06	0.18	0.6732
QPec vs. AT	0.12	1	0.12	0.36	0.5501

## Contrast coefficients

Location	Cont.1	Cont.2
Arroyo del Tanque	1.00	-1.00
Maricao Forest Reserve	-4.00	0.00
Quebrada Calliandra	1.00	0.00
Quebrada Grande	1.00	0.00
Quebrada Peces	1.00	1.00

## Test:LSD Fisher Alpha=0.05 DMS=0.58525

Error: 0.3393 df: 102			
Location	Means	n	
Quebrada Peces	0.34	47	А
Arroyo del Tanque	0.27	51	А
Quebrada Grande	0.19	2	А
Maricao Forest Reserve	0.07	б	А
Quebrada Calliandra	-0.03	1	А
Different letters indicate significan	nt difference	es (p<= 0.	05)

	T 1'.
Planted by	Locality
Miguel A."Papo" Vives-Heyliger	Quebradillas, in front of his residence and at the gardens of some of his neighbors.
Felipe Osbourne	In Mr. Osbourne's plantation (Machabuca); Mr. Vives provided the seeds.
Rubén Padrón-Vélez	Maricao Forest Reserve at the old camping area, at Quebrada Negra and in front of the DNER offices. At the Guilarte Forest Reserve in front of the DNER offices, and in front of his residence in Yauco, PR
DNER personnel	Quebrada Piedras, Maricao Forest Reserve, 21 plants by the resident forest management official's house and 43 upstream west of an old water tank.
DNER personnel	Between 75 and 120 plants were planted at the DNER's Finca Gabia and Toa Vaca in Coamo, PR
Susan Silander and Miguel Canals	In front of the resident forest manager official's house at the Guánica Forest Reserve.
Carlos Figueroa-Colón	UPRM, west of the Carlos E. Chardón building.
Botanical garden personnel	Caguas Botanical Garden in Caguas, PR Seven plants, five appear to be hybridizing.
Garden's personnel	In plots 29, 61, 112, 158.

Appendix VI. Localities for cultivated plants of *Crescentia portoricensis* Britton.

Nursery personnel	Fairchild Tropical Garden in Miami, FL. One hundred plants in pots, at the DNER's Cambalache Forest Reserve nursery in Arecibo, PR
Nursery personnel	One hundred plants in pots in the Conservation Trust of Puerto Rico's nursery in Rio Piedras, PR
Reserve personnel	Eighty-six plants in pots in the Conservation Trust of Puerto Rico's, Cañón de San Cristóbal in Barranquitas, PR