Survey for potential biological control agents for

Miconia calvescens in southern

Mexico: final report

Prepared for the Hawaii Invasive Species Council

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1 Summary

A two-week survey was conducted in August 2006, in the state of Chiapas, Mexico, to locate populations of *Miconia calvescens* and to identify insect and other natural enemy species present on it that might have potential as biological control agents. M. calvescens was found to be present in Chiapas, although at very low population densities. Seven locations were found in two separate areas, one covering parts of the municipalities of La Independencia and Maravilla Tenejapa, east of the Lagunas de Montebello National Park, and the other in the municipality of Ocosingo, near the Metzabok Natural Area. All locations of *M. calvescens* were in shaded secondary roadside vegetation between about 450 m and 1,100 m elevation. All plants were in the vegetative stage except for one that had flower buds. A number of insect species were collected, including larvae of Riodinidae, Geometridae, and Megalopygidae (Lepidoptera). Herbarium specimens and DNA samples of *M. calvescens* were collected at all sites. Plant and insect material collected was shipped from Mexico to the cooperating laboratory at the University of Costa Rica. A habitat analysis was conducted to identify further areas in the state of Chiapas that might be suitable habitat for *M. calvescens*. Support for the survey was provided by ECOSUR, a Mexican research institution which could be a valuable partner for further studies on potential biological control agents for *M. calvescens*. It is recommended that further surveys of areas identified as potential habitat for M. calvescens should be conducted at other times of year to locate additional populations and to further document its natural enemies. Planting cohorts of M. calvescens seedlings in areas where it occurs naturally could provide useful information on the role of natural enemies in its population dynamics, and assist in identifying suitable biological control agents.

2 Background and objectives

Miconia calvescens is a shrub or small tree in the family Melastomataceae, native to rainforests of tropical America, which has become a major invader of native forest ecosystems in several Pacific island groups. It has become dominant over 65% of the island of Tahiti, displacing native forest communities and threatening up to 100 native plant species (Meyer and Florence 1996). *M. calvescens* was found to have escaped from cultivation on Maui in 1990, and efforts to contain established populations have been under way since then. Invasive populations also have been targeted for control on Hawaii, Oahu and Kauai. It is considered to represent a threat to all habitats in Hawaii receiving more than 1800 mm annual precipitation (Loope 1997).

Studies on potential biological control agents for *M. calvescens* were initiated in 1993 with surveys in Costa Rica, Brazil, Uruguay, Paraguay, Argentina, and Trinidad and Tobago. Currently the US Forest Service is coordinating studies of several potential biocontrol agents in Costa Rica and Brazil (Killgore et al. 1999; Santos-Seixas et al. 2002; Santos-Seixas et al. 2004; Burckhardt et al. 2005; Badenes-Perez and Johnson 2007a; Badenes-Perez and Johnson 2007b).

The native range of *M. calvescens* extends from Brazil to southern Mexico (Wurdack 1980). According to Meyer (1998), Mexico was the source of *M. calvescens* plants cultivated at the Peradeniya Botanic Gardens in Sri Lanka, from where they were introduced to Tahiti and probably to other Pacific islands. Thus, Mexican populations of *M. calvescens* may be the most closely matched to the invasive biotypes in the Pacific islands, making them particularly suitable as a source of biocontrol agents. However, no surveys had previously been conducted of the natural enemies of *M. calvescens* in Mexico.

The goal of this project was to survey the insects, mites, nematodes and pathogens associated with *M. calvescens* at sites where it has been reported to occur in the Mexican states of Chiapas and Oaxaca. The objectives were to determine:

- 1. Are there substantial populations of *M. calvescens* in southern Mexico that could harbour natural enemies?
- 2. Are *M. calvescens* populations in Mexico morphologically and genetically similar to the invasive populations present in Hawaii and other Pacific islands?
- 3. Are there additional natural enemy species present in these populations that would expand the pool of potential biocontrol agents beyond that currently known from Brazil, Ecuador, and Costa Rica?
- 4. Are local collaborators and facilities available for further studies on potential biocontrol agents?
- 5. Would further studies based in Mexico be warranted and what costs would be involved?

3 Survey methodology

Areas for survey were selected on the basis of known records for *M. calvescens* in southern Mexico. From online herbarium records at the California Academy of Sciences (CAS), *M. calvescens* sites had previously been identified in the municipalities of La

Trinitaria, Ocosingo, and Palenque (state of Chiapas) and Santa María Chimalapa (state of Oaxaca). The REMIB database (Red Mundial de Información sobre Biodiversidad) at <u>www.remib.com</u> was searched for additional Mexican records. Immediately before starting this survey, I visited the Mexican National Herbarium (MEXU) in Mexico City to examine their *Miconia* holdings for other sites of *M. calvescens*. Photographs taken of herbarium specimens at MEXU and other images from Internet sources were used to produce a field guide to *Miconia* species in Chiapas that was useful in tentatively identifying some other species found during the survey. Names of other *Miconia* species mentioned in this report are strictly tentative visual identifications based on comparison with these images and others obtained from Internet sources, and are not authoritative.

The survey was essentially a roadside survey, with some exploration on foot in areas where *M. calvescens* was found or had been reported. On the basis of known sites, a route was planned starting in Tuxtla Gutiérrez, Chiapas, and proceeding southeast along Mexican Highways 190 and 307 to the Lagunas de Montebello National Park; following Highway 307, which loops around the eastern end of the state of Chiapas, to the Bonampak area; cross-country on back roads to Ocosingo; north on Highway 199 to Palenque; continuing north to Highway 186 and briefly entering the state of Tabasco before turning south again on Highway 195 through Teapa and back to Tuxtla Gutiérrez. For the last two days the area west of Tuxtla Gutiérrez from Ocozocoautla to Presa Nezahualcoyotl and Chicoasén was explored (Figure 1). We did not visit the reported site at Santa María Chimalapa, Oaxaca, due to security concerns in Oaxaca. A full itinerary is provided in APPENDIX II.

At all sites where *M. calvescens* was encountered, herbarium specimens were taken and shoot tip material was preserved dried in silica gel for DNA analysis. Any insects found associated with *M. calvescens* were collected. Adult insects were pinned or preserved in 95% ethanol, and larval stages were collected alive for rearing through to adults where possible. Some collections were also made on other *Miconia* species. All sites were documented by digital photography, and routes and collecting sites were located by GPS using a Garmin GPSmap 60CS receiver.

I was ably accompanied and assisted on this survey by Dr. Benigno Gómez, an entomologist from El Colegio de la Frontera Sur (ECOSUR), Tapachula, Chiapas, and Dr. Carlos Beutelspacher, retired entomologist and botanist at the National Autonomous University of Mexico (UNAM) and former director of the Chiapas state Institute of Natural History (IHN).

4 Results

4.1 Herbarium records

The National Herbarium in Mexico City was found to have relatively small holdings of *Miconia* species from Chiapas, although there is much material from other southern Mexican states such as Oaxaca and Veracruz. A specimen originally labelled as *Miconia impetiolaris* and later reidentified as *M. calvescens* was labelled "Cafetal Irlanda, Huixtla, Chis. 600 m. 26.XII.1968". This location is in southern Chiapas, near Tapachula, and was not on our survey route. One record dated October 7, 1992, in the REMIB database shows *M. calvescens* at Solosuchiapa, Chiapas: the record does not indicate whether this refers to the town or the municipality of Solosuchiapa. This is on Highway 195, north of

Tuxtla Gutiérrez, and was included in our survey route. Two records of *M. calvescens* from Santa María Chimalapa, Oaxaca, were duplicates of sites already known from the CAS database. A very old record (May 30, 1939) was also found in the REMIB database for Atoyac de Álvarez, in the state of Guerrero.

After my return I received a digital image of the *M. calvescens* specimen from Solosuchiapa, courtesy of Dr. Jerzy Rzedowski at the herbarium of the Instituto de Ecología del Bajío (IEB). The label information indicates that the specimen was collected in "bosque tropical perennifolio" at an elevation of 430 m.

4.2 Survey itinerary

The survey itinerary is shown in Figure 1.

Aug. 10 2006. Arrived in Mexico City by air from Edmonton via Phoenix.

Aug. 11, 2006. Visited National Herbarium (MEXU).

<u>Aug. 12, 2006</u>. Arrived by air in Tuxtla Gutiérrez, capital of the state of Chiapas, rented a vehicle (Jeep Liberty from Hertz), and met with Benigno Gómez, entomologist with ECOSUR. We met later with Dr. Carlos Beutelspacher who was to accompany us on the survey, and purchased food and other supplies.

<u>August 13, 2006</u>. Drove from Tuxtla Gutiérrez via San Cristóbal de las Casas and Comitán to the Lagunas de Montebello National Park, arriving in the late afternoon. We met with staff of the Comisión Nacional de Areas Naturales Protegidas (National Commission for Protected Natural Areas: CONANP) and spent the night in the rather rustic visitor accommodation at the CONANP park headquarters.

<u>August 14, 2006</u>. Accompanied by a local CONANP park ranger, Roberto Castellanos, we explored areas within and near the Lagunas de Montebello National Park. We showed photographs of *M. calvescens* to Sr. Castellanos and other local residents who recognized the plant and said that it is found along river edges but were not able to report exact locations. According to Sr. Castellanos the local name for the plant is "tilinté"; he indicated that there is another species also with a red underside to the leaf but with smaller leaves – from this description this may be *Miconia oinochrophylla*. We surveyed areas around Lago Montebello and along a road and a riparian area leading north out of the National Park towards Tierra Blanca at elevations of around 1500 to 1300 m. Other Melastomataceae and *Miconia* species were seen but no *M. calvescens*. In the afternoon we surveyed along the road leading east from the National Park (highway 307) and found single plants of *M. calvescens* at three locations around 12 km NW of Tziscao at elevations between 1135 and 985 m (Figure 2), and a further plant along this same road about 5 km SW of Amparo Agua Tinta at an elevation of 838 m (Figure 3). Returned to the CONANP headquarters at Lagunas de Montebello for the night of August 14.

<u>August 15, 2006</u>. Drove east along highway 307 from Lagunas de Montebello, with a side trip of about 20 km along a road running NW from El Carmen along the Rio Santo Domingo to Esmeralda, returning by the same route to 307. Another *Miconia* species (possibly *M. dodecandra*?) was seen near Gracias a Dios along this road but no *M. calvescens*. Collected DNA samples of *M. calvescens* at same sites located the previous

day. Continuing east along 307 we arrived at the Centro Ecoturístico Las Nubes, located about 6 km N of route 307 along an access road starting near Jerusalén. *Miconia impetiolaris* was seen along a riverside trail at Las Nubes and two other species, possibly *M. serrulata* and *M. argentea*, were noted along the access road to Las Nubes.

<u>August 16, 2006</u>. Surveyed on foot along river shore in vicinity of Las Nubes. A small *Miconia* species seen (possibly *M. lacera*) but no *M. calvescens*. Returned to highway 307 and continued east where we located another *M. calvescens* plant near Maravilla Tenejapa at an elevation of 483 m (Figure 4). Immature flower buds were seen on this plant (Figure 5) Continued east into flatter, low-lying country along the Río Lacantún, and took the road north through Chajul towards Pico de Oro heading for the San Javier/Bonampak area. This road turned out to be blocked by construction about 30 km north of the turn-off, so we returned a few km to the Centro Ecoturístico Las Guacamayas where we spent the night.

<u>August 17, 2006</u>. Returned from Las Guacamayas south to highway 307. Several plants of a tall *Miconia* sp. similar in habit to *M. calvescens* but with leaves brown on the underside (possibly *M. elata*) were found just inside forest cover a few km south of Chajul (Figure 6). Rejoined 307 and continued initially eastwards. This road loops around the eastern end of the Marqués de Comilla region, an area that has been heavily deforested and largely converted to pasture. Continued through Benemérito de las Américas and northwest towards San Javier, stopping at another reported location of *M. calvescens* "14 km NW Crucero Corozal". Habitat appeared suitable for *M. calvescens* (Figure 7) but none was seen in a search along about 500 m of the road. One small plant of *M. impetiolaris* was seen. We continued on to the CONANP offices at Lacanjá Chansayab where we met with CONANP staff and local Lacandón residents and spent the night.

<u>August 18, 2006</u>. Walked with a local Lacandón guide from Lacanjá Chansayab along forest trails to Ya Toch Kusam waterfalls and a small Mayan ruin site. No *M. calvescens* was seen but *M. impetiolaris* was fairly common (Figure 8). This species and another, possibly *M. dodecandra*, were also common along the road to the Bonampak ruins. We continued approximately another 65 km NW along highway 307 and stopped for the night at the Hotel Camino Verde near Nuevo Canán.

<u>August 19, 2006</u>. Drove NW along highway 307 as far as Chancalá and then turned south and east towards Metzabok Natural Area. Two plants of *M. calvescens* were found near the access road to Metzabok Lake at elevations of 577 and 644 m (Figure 9). We also surveyed around Nahá Lake in the Nahá Natural Area in similar habitats but no *M. calvescens* were found. Continued south to Monte Líbano and eastwards on rough roads to Ocosingo. Much of this country has been heavily deforested, at higher elevations up to 1183 m the road passes through pine forest.

<u>August 20, 2006</u>. Drove Ocosingo to Palenque along highway 199, few *Miconia* spp. seen. Surveyed on foot at Agua Azul and Misol-Há waterfalls, the latter being an area where *M. calvescens* had been reported, but none was seen. Intermittent heavy rain later in the day, the first rain encountered on the survey except for thunderstorms at night.

August 21, 2006. Rest day in Palenque for laundry, email, etc.

<u>August 22, 2006</u>. Drove from Palenque north on highway 199 to highway 186, west towards Villahermosa, crossing into the state of Tabasco, then south through Jalapa and Teapa, returning into Chiapas through Pichucalco and south on highway 195. The earlier parts of this route are low-lying coastal plain and largely agricultural, with little suitable habitat for *Miconia*. Stopped at the townsite of Solosuchiapa where *M. calvescens* had been reported (although the exact location of the record is not clear) and searched on foot along the riverbank area (Figure 10) but no *M. calvescens* was seen. Continued south towards Tuxtla Gutiérrez on 195. Habitat for around 20 km south of Solosuchiapa appeared suitable for *Miconia* but none was seen. Further south this road passes through areas of high pine forest, up to around 1800 m around Tapilula, before descending to Chiapa de Corzo and Tuxtla Gutiérrez where we arrived about 7 p.m.

<u>August 23, 2006</u>. Surveyed along the new highway NW of Tuxtla Gutiérrez towards the Presa Netzahualcoyotl reservoir, across the reservoir and returning to Tuxtla via Tecpatán, Copainalá and Chicoasén. Several other *Miconia* species were seen including possibly *M. chrysophylla* and *M. impetiolaris*, but no *M. calvescens*, although the habitat appeared suitable in some areas, particularly in forest remnants at around 930 m elevation SW of the Chicoasén reservoir (Figure 11).

<u>August 24, 2006</u>. Surveyed in hilly areas up to 10 km NW of Berriozábal at elevations of 900 – 1200 m: coffee plantations with fields and forest remnants, and a forested area known as "El Pozo". Habitat appeared suitable but no *M. calvescens* were seen. Returned to Berriozábal and surveyed along the "old road" from Ocozocoautla towards Presa Netzahualcoyotl. A roadside location near "Laguna Bélgica" at 968 m elevation had two smaller shrubby *Miconia* species on which we collected lepidoptera larvae and apparently nematode galls. Continued along this road to where it joins the new highway to Presa Netzahualcoyotl and then returned along the same route to Ocozocoautla, Berriozábal and Tuxtla Gutiérrez.

<u>August 25, 2006</u>. Partial rest day. Visited Cañon del Sumidero NE of Tuxtla Gutiérrez, natural vegetation in good condition at elevations of 900 - 1300 m but no *Miconia* species seen. Packed insect and plant collections ready for further rearing and shipping and handed them over to Benigno Gómez who transported them to ECOSUR at Tapachula by overnight bus. Packed ready for return to Canada.

<u>August 26, 2006</u>. Returned rental vehicle at Tuxtla Gutiérrez airport and flew Tuxtla – Mexico City – Phoenix – Edmonton.

4.3 Miconia calvescens populations

M. calvescens was found in two areas of Chiapas, east of the Lagunas de Montebello National Park, and near the Metzabok Natural Area. Two locations along Highway 307 just east of the Lagunas de Montebello National Park are probably close to those from which *M. calvescens* was collected by Breedlove in 1982 (specimens CAS 673154 and 674626). *M. calvescens* was found at another site about 32 km east of these sites along Highway 307, near Maravilla Tenejapa. Two plants were found about 2 km apart on unnamed roads just outside the boundaries of the Metzabok Natural Area. We were not able to confirm the records of *M. calvescens* from the Bonampak area (San Javier or Crucero Corozal), the Misol-Há waterfall (about 20 km south of Palenque) or Solosuchiapa (see Figure 33). A full list of sites where *M. calvescens* was found, as well as other sites in southern Mexico from which it has been reported, is given in Table 1.

M. calvescens was found in shaded secondary roadside vegetation between about 450 m and 1,100 m elevation. It was never seen in open areas or under dense forest canopy. Population densities were extremely low. A total of 7 sites were found, most consisting of single plants, ranging in height from 1 to 8 m. All plants were vegetative except for one that had immature flower buds. No seedlings of *M. calvescens* were seen at any site.

4.4 Insects and other potential agents found

Most insects found were foliage-feeding Lepidoptera. An adult *Euselasia* sp. (Riodinidae) was reared out from a larva collected in the Lagunas de Montebello area (Figure 12, Figure 13, Figure 14, Figure 15, Figure 16). This belongs to a genus in which other members have already been studies as possible biological control agents for *M. calvescens*. A group of gregariously-feeding larvae found at this site (Figure 17, Figure 18) may have been of the same species but stopped feeding before they could be reared through to adults. After my departure another riodinid adult emerged from one of the larvae left at ECOSUR.

A number of other foliage-feeding Lepidoptera larvae were found, including species of Megalopygidae, Geometridae, and other families (Figure 19, Figure 20, Figure 21, Figure 22, Figure 23, Figure 26). Some of these pupated after collection, and adults may be reared out for identification.

Leaves with extensive feeding damage were sometimes found without any signs of what had caused the damage (Figure 24). Some leaves showed rasping damage (Figure 25) similar to that caused by *Atomacera* sp. (Badenes-Perez and Johnson 2007a). At one site dead petiole bases were seen on some stems (Figure 27), suggesting damage by some type of internally feeding insect. However, no larvae or definite signs of insect damage were seen in these petioles.

Cottony filaments on shoot tips suggesting the possible presence of immature psyllids were collected at the Metzabok site.

At one site near Laguna Bélgica, northwest of Tuxtla Gutiérrez, a small *Miconia* species, possibly *M. chrysophylla*, was attacked by a number of species similar to others that have been found on *M. calvescens*. A lepidopteran larva (Figure 28) was similar to larvae collected on *M. calvescens* (Figure 26). Massive, spongy leaf galls (Figure 29) closely resembled those caused by nematodes that have been found on *M. calvescens* in Costa Rica and Brazil. Larvae feeding on the leaf surface caused similar damage to that attributed to *Atomacera* (Figure 32). Cottony filaments emerging from shoot tips (Figure 30) were similar to those produced by *Diclidophlebia lucens* on *M. calvescens* (Burckhardt et al. 2005). Another possible psyllid laid eggs in lines along the veins on the underside of leaves of the large *Miconia* (possibly *M. elata*) found near Chajul (Figure 31).

Few obvious signs of disease were seen on M. calvescens but some leaf spots were observed on mature leaves on the plant near Maravilla Tenejapa (Figure 32).

All insect and plant material collected was transported to ECOSUR headquarters at Tapachula by Benigno after my departure and sent by DHL courier to Dr. Paul Hanson at the University of Costa Rica on March 13, 2007. Unfortunately this shipment disappeared in transit for a considerable time before it was finally confirmed that it was awaiting clearance from Costa Rican customs in October 2007. As of September 2008 it appears that the shipment has not yet been retrieved from customs.

5 Predicted habitats for Miconia calvescens

Based on the sites where *M. calvescens* was found during this survey in Chiapas, an attempt was made to predict other areas of the state where it would be likely to occur. GIS data for the state of Chiapas were provided by the Laboratorio de Análisis de Información Geográfica y Estadística (LAIGE) at ECOSUR, and digital elevation data from the Seamless Shuttle Radar Topography Mission (SRTM) "Finished" 3 Arc Second dataset (90 m resolution approximately) were downloaded from the US Geological Survey's Seamless Server site at <u>http://seamless.usgs.gov/index.php</u>. Based on the data for confirmed and reported sites of *M. calvescens*, the following characteristics were selected: vegetation "Selva alta y mediana perennifolia" or "Selva alta y mediana perennifolia con vegetación secundaria arbustiva y herbacea"; soil "Rendzina, Feozem, or Acrisol"; climate type (A)C(m), Am, or Af(m); elevation 450 – 1150 m (see Table 2). Sites at Metzabok were classified as cultivated pasture, but were within 1 km of areas classified as "Selva alta y mediana perennifolia con vegetación secundaria arbustiva y herbacea". Rather than including pasture as a habitat for *M. calvescens*, a 1-km buffer was therefore added around all areas of the first two types of vegetation.

Areas predicted to be suitable for *M. calvescens* on the basis of this analysis are shown in purple in **Figure 33**. Three main areas are shown as including potentially suitable habitat:

- 1. Eastern areas of the state from the Guatemalan border north to around the Metzabok Natural Area. Much of this area lies within the Montes Azules Biosphere Reserve, a largely pristine and roadless area where access would be difficult. Areas west of the Biosphere Reserve would be accessible by back roads from Comitán.
- 2. Scattered areas in the north-western part of the state, along the borders with Tabasco, Veracruz and Oaxaca. Much of this area is cleared for agriculture but suitable *M. calvescens* habitat may remain in forest remnants or in protected areas such as the Selva El Ocote Biosphere Reserve.
- 3. A relatively small area in the El Triunfo Biosphere Reserve in the southern part of the state. Much of this reserve is still in good ecological conditions and access by road and trails is good.

6 Conclusions and Recommendations

Miconia calvescens has been confirmed by this survey to occur in Chiapas, although at very low densities. Further searches in the vicinity of the sites identified on this survey, and in the areas of potentially suitable habitat identified in the GIS analysis (Figure 34) should lead to more locations of *M. calvescens*. The rarity of *M. calvescens* in Chiapas, in comparison with its high density and rapid spread in invaded Pacific islands, suggests

that environmental factors, possibly including natural enemies such as insects and plant diseases, limit its populations in this area of its native range.

ECOSUR (El Colegio de la Frontera Sur, <u>www.ecosur.mx</u>) is well placed to act as a cooperating agency for further studies on potential biological control agents for M. calvescens in southern Mexico. ECOSUR is a publicly funded, multidisciplinary institute for research and postgraduate education, focussing on the sustainable development of the southern regions of Mexico. Its headquarters is in Tapachula (Chiapas), and it has campuses in San Cristóbal de las Casas, Chetumal, Campeche, and Villahermosa. Its areas of research include the conservation of biodiversity, alternative production systems, agroecology, management of natural resources, and health and socioeconomic issues in the region. The staff includes entomologists and botanists and the organization has an insect collection, herbarium and botanic garden, as well as libraries, internet access, and a range of laboratory facilities including GIS support. ECOSUR researchers are widely recognized in Mexico and internationally; in 2006 researchers published 122 scientific articles, 31 book chapters and 3 books, and in the same period graduate students at ECOSUR completed 13 doctoral and 54 masters' theses (ECOSUR 2007). Much of their research is conducted at field sites in natural and managed ecosystems throughout southeastern Mexico, and they are able to obtain the required permits for field collection and research from appropriate authorities in Mexico. The San Cristóbal and Tapachula campuses are well located for access to areas where M. calvescens is expected to occur. The director-general of ECOSUR, Dr. Pablo Liedo Fernández, is himself an entomologist and was extremely helpful and supportive in the planning of this survey, as were all ECOSUR staff that I dealt with.

My recommendations for further study of potential biological control agents for *M. calvescens* in Mexico are:

1. Further surveys should be conducted in the areas of Chiapas identified as potential habitat for *M. calvescens* in this report, to locate additional populations and further document the natural enemy complex associated with this species.

2. The current survey was conducted during the rainy season. Future surveys should include field work at other times of year to ensure that insect and other natural enemy species with varying phenologies are detected. In general the rainy season in Chiapas runs from approximately June to October.

3. Discussions should be held with ECOSUR on the possibility of initiating graduate student research projects on the ecology of *M. calvescens* and its natural enemies.

4. Field experiments using planted cohorts of *M. calvescens* seedlings in areas where it occurs naturally may be valuable in understanding its population dynamics in this part of its native range, as well as in recruiting populations of natural enemy species for further study.

5. Natural enemy complexes associated with other *Miconia* species occurring in Chiapas should be investigated to provide additional information on the host-specificity of potential biological control agents.

6. DNA studies on material of *M. calvescens* from Mexico and other areas of its native range, as well as invasive Pacific populations, should be continued to identify the most likely origin of the invasive populations and to aid in selecting the most best-matched biological control agents for these.

7 Acknowledgments

This survey would not have been possible without the assistance of M. en C. Benigno Gómez, ECOSUR, both in the planning and execution stages. He assisted with route planning, identification of field sites, obtaining collecting permits, making contact with local national park staff and guides, field collection, and rearing out of live material. I am also very grateful to Dr. Carlos Beutelspacher for accompanying us on the survey and for the benefit of his deep knowledge of the flora, fauna, natural areas, and back roads of Chiapas. I am also grateful to Dr. Gerardo Salazar for assistance at the National Herbarium of Mexico, to Dr. Frank Almeda of the California Academy of Sciences for information on *M. calvescens* locations, and to Dr. Jerzy Rzedowski for the image of *M*. calvescens from IEB. Sr. Roberto Castellanos of the National Commission of Protected Natural Areas (CONANP) was a knowledgeable local guide at the Lagunas de Montebello National Park, and Sr. Mario Chan Bor guided us along rain forest trails at Lacanjá Chansayab, near Bonampak. Dr. Tracy Johnson, US Forest Service, assisted with planning the survey and provided information on ongoing biological control work with M. calvescens. Alejandro Flamenco Sandoval of the Laboratorio de Análisis de Información Geográfica y Estadística at ECOSUR kindly provided GIS data on the vegetation, soils, and climates of Chiapas. Particular thanks are due to Dr. Pablo Liedo Fernández, director-general of ECOSUR, for facilitating collaboration by ECOSUR staff in this survey.

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Longitude °W	Latitude °N	Elevation (m)	Date and time	Location	Herbarium data	Notes
91.5641	16.1108	1135	Aug 14 2006, 12:46 pm	Municipio La Trinitaria	CAS 674626, 10 km ENE of Dos Lagos above Santa Elena, Coll. Breedlove 1982	Confirmed in this survey
91.5601	16.1162	994	Aug 14 2006, 1:58 pm	Municipio La Trinitaria	CAS 673154, 15 km ENE of Dos Lagos above Santa Elena, Coll. Breedlove 1982	Confirmed in this survey
91.5600	16.1168	985	Aug 14 2006, 2:17PM	Municipio La Trinitaria	CAS 673154, 15 km ENE of Dos Lagos above Santa Elena, Coll. Breedlove 1982	Confirmed in this survey
91.2609	16.1381	483	Aug 16 2006, 12:45 pm	near Maravilla Tenejapa		New record
91.6367	17.1031	577	Aug 19 2006, 12:01 pm	Metzabok Natural Area		New record
91.6474	17.0949	644	Aug 19 2006, 12:52 pm	Metzabok Natural Area		New record
91.0939	16.8011	411	Aug 17 2006, 3:02 pm	14 km NW Crucero Corozal	NYBG Specimen ID: 5212, coll. 1986	Reported, not found
91.9996	17.3919	247	Aug 20 2006, 5:08 pm	Misol-Há waterfall	CAS 670513, Near Cascada Mizola, 25 km S of Palenque on road to Ocosingo	Reported, not found
93.0279	17.4257	430	Aug 22 2006, 1:57 pm	Solosuchiapa (Coordinates are for the town of Solosuchiapa – the locality record may refer to the municipality, which covers a much larger area)	IEB 88959, Coll. 1992	Reported, not found.
92.3333	15.1667	600		Cafetal Irlanda, Huixtla, Chiapas	MEXU, coll. 1968 Miconia cf. calvescens	Not on the survey route.
94.7333	16.8833	250 - 300		Santa Maria Chimalapa, Oaxaca	MEXU, coll. 1985	Not on the survey route.
100.3747	17.3986	unknown		Plan del Carrizo, Atoyac de Álvarez, Guerrero	LL266723 coll. May 30, 1939	Not on the survey route.

Table 1. Locations at which *Miconia calvescens* was found in Chiapas in the survey, and other locations in southern Mexico from which it has been reported.

Fields	Values from LAIGE database	Notes/translation
Vegetation	"Selva alta y mediana perennifolia"	Tall and medium evergreen forest
	"Selva alta y mediana perennifolia con vegetación secundaria arbustiva y herbacea"	Tall and medium evergreen forest with secondary woody and herbaceous vegetation.
	"Pastizal cultivado"	Cultivated pasture.
Soil	Rendzina, Feozem, Acrisol	Rendzina, Phaeozem, or Acrisol
Climate		[According to Köppen classification as modified by García (Soto-Esparza et al.)]
	(A)C(m) Subgrupo semicálido proveniente del grupo de climas C. La temperatura media anual es mayor de 18°C, la media del mes más frío está comprendida entre -3 y 18°C y la media del mes más caliente superior a 6.5°C. Porcentaje de lluvia invernal entre 5 y 10.2.	(A)C(m) "Semi-hot" subgroup belonging to the C (temperate) group. Mean annual temperature above 18°C, mean of the coldest month between -3 and 18°C and mean of the hottest month above 6.5°C. Percentage of rain in winter between 5 and 10.2.
	Am Grupo cálido. Temperatura media anual entre 22 y 26°C ó mayor a este último valor. La temperatura media del mes más frío es superior a 18°C. Régimen de lluvias de verano con influencia de monzón. El porcentaje de lluvia invernal está comprendido entre 5 y 10.2.	Am. "Hot group". Mean annual temperature between 22 and 26°C or higher. Mean temperature of the coldest month above 18°C. Summer rainfall regime with monsoon influence. Percentage of winter rain between 5 and 10.2
	Af(m) Grupo cálido. Temperatura media anual entre 22 y 26°C ó mayor a este último valor. La temperatura media del mes más frío es superior a 18°C. Régimen de lluvias todo el año; el mes más seco con precipitación mayor a 60 mm. El porcentaje de lluvia invernal es menor de 18.	Af(m) "Hot group". Mean annual temperature between 22 and 26°C or higher. Mean temperature of the coldest month above 18°C. Year-round rainfall regime; driest month with at least 60 mm precipitation. Percentage of winter rain below 18.
Elevation	Between 450 and 1150 m.	Actual lowest and highest elevations at which <i>M. calvescens</i> was found in the survey were 483 and 1135 m.

Table 2. Site characteristics used in predicting suitable habitat for *Miconia calvescens* in Chiapas.

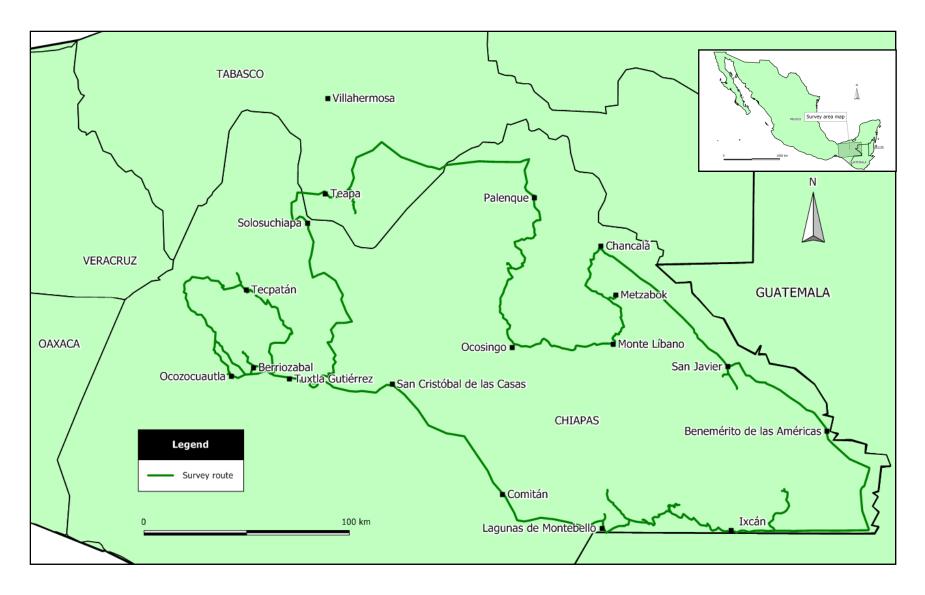


Figure 1. Miconia calvescens survey route and sites in southern Mexico.



Figure 2. *Miconia calvescens* plant in roadside vegetation about 12 km NW of Tziscao.



Figure 3. Larger *M. calvescens* plant 5 km SW of Amparo Agua Tinta.



Figure 4. B. Gómez and C. Beutelspacher collecting on *M. calvescens* plant near Maravilla Tenejapa.



Figure 5. Flower buds on *M. calvescens* near Maravilla Tenejapa.



Figure 6. Large *Miconia* species near Chajul, possibly *M. elata*.



Figure 7. Reported locality for *M. calvescens* near Crucero Corozal.



Figure 8. *Miconia impetiolaris* in forest understorey near Lacanjá Chansayab.



Figure 9. M. calvescens near Metzabok Natural Area.



Figure 10. Río La Sierra near Solosuchiapa.



Figure 11. Forest remnant near Chicoasén.



Figure 12. Larva of Euselasia sp. collected on M. calvescens

Figure 13. Pupa of Euselasia sp.



Figure 14. Adult *Euselasia* sp. emerged from the pupa in the previous figure.



Figure 15. Adult *Euselasia* sp., pinned, upper side.



Figure 16. Adult *Euselasia* sp., pinned, underside.



Figure 17. Gregarious lepidoptera larvae on *M. calvescens*, possibly also *Euselasia* sp.



Figure 18. Same larvae as in the previous figure, after a few days feeding.

Figure 19. Lepidoptera larva on M. calvescens.



Figure 20. Lepidoptera larva on *M. calvescens*.

Figure 21. Geometrid larva on M. calvescens.



Figure 22. Lepidoptera larva on *M. calvescens*, probably *Megalopyge* sp.



Figure 23. Lepidoptera larva on M. calvescens.



Figure 24. Feeding damage by an unknown agent on *M. calvescens*.

Figure 25. Rasping damage similar to that caused by *Atomacera* spp. on *M. calvescens*.



Figure 26. Lepidoptera larva on M. calvescens.

Figure 27. Withered petiole of *M. calvescens*.



Figure 28. Lepidoptera larva on Miconia sp. possibly chrysophylla.



Figure 29 Galls on Miconia sp. possibly chrysophylla.



Figure 30. Shoot tip of Miconia sp. possibly chrysophylla with
possible psyllid infestation.Figure 31. Eggs and nymphs of Homoptera (psyllids?) on
foliage of Miconia sp. possibly elata.



Figure 32 Sawfly? larvae feeding on leaves of *Miconia* sp. possibly *chrysophylla*.

Figure 33 Possible fungal leaf spot on foliage of *Miconia* calvescens near Maravilla Tenejapa.

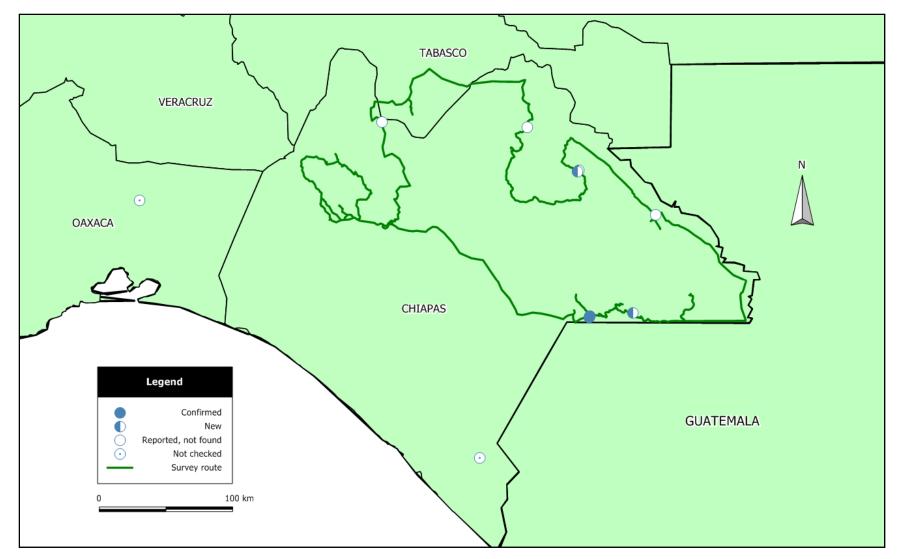


Figure 34. Confirmed and reported locations of *Miconia calvescens* in southern Mexico.

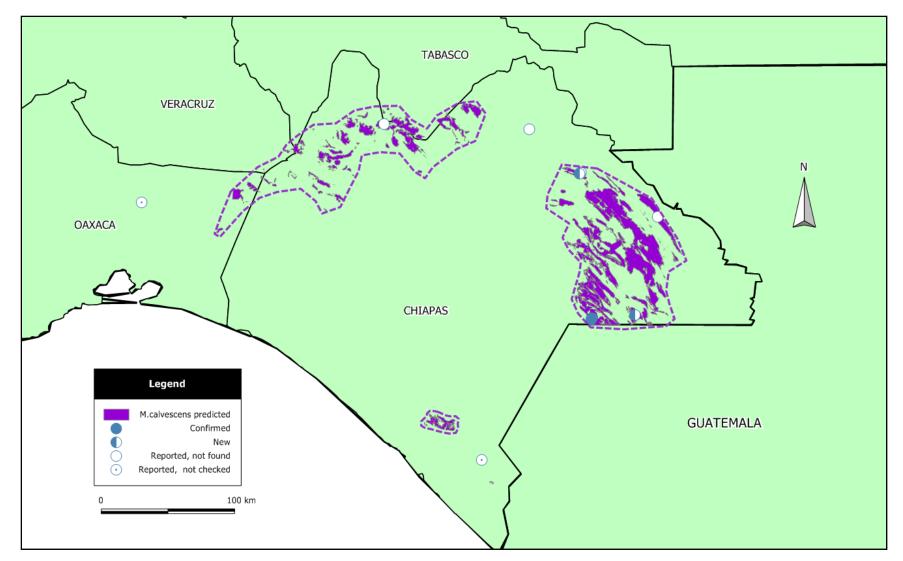


Figure 35. Areas of Chiapas predicted to be suitable for *M. calvescens* on the basis of a GIS analysis of soil, vegetation, climate and elevation.

APPENDIX I: List of material

Material from *Miconia calvescens* survey in Chiapas, Aug. 12-26 2006, left with Dr. Benigno Gómez, ECOSUR, and subsequently shipped to Dr. Paul Hanson, UCR.

Herbarium specimens

Temporary label	Data
Aug. 14 1:30 p.m.	<i>Miconia calvescens</i> . (3 samples.)
[this is WP47]	Roadside. No flowers, buds or fruit.
	Collected Aug. 14, 2006. N16.11084 W91.56408, alt. 1135 m.
WP50	Miconia calvescens.
	Slender, single-stemmed, branching tree, c. 6 m. high.
	In shade near roadside. No flowers, buds or fruit.
	Collected Aug. 14, 2006. N16.11162 W91.45116, alt. 838 m.
WP58	Miconia calvescens.
	Small multi-stemmed tree c. 7 m. high. In shade between roadside and
	banana plantation. Flower buds present.
	Collected Aug. 16 2006, N16.13808 W91.26092, alt. 483 m.
WP60	Melastomataceae?
	Small tree, flowers white, fragrant. Common along roadsides in this area.
	Collected Aug. 16 2006, N16.08325 W90.98571, alt. 218 m.
WP62	Miconia unidentified sp.
	One of 6 small trees $2 - 12$ m. high. No flowers, buds or fruit.
	West of road, just inside forest cover.
	Collected Aug. 17 2006, N16.11165 W90.92133, alt. 180 m.
WP68	Miconia calvescens.
[should read	Multi-stemmed small tree c. 6 m. high. In shade by roadside. No flowers,
WP69]	buds or fruit.
	Collected Aug. 19, 2006. N17.10310 W91.63666, alt. 577 m.
WP70	Miconia calvescens.
	Single-stemmed tree c. 8 m. high. In shade between road and coffee
	plantation. No flowers, buds or fruit.
	Collected Aug. 19, 2006. N17.09490 W91.64741, alt. 644 m.
WP97	Miconia unidentified sp.
	Small bush c. 1 m high. Roadside at base of rocky outcrop. No flowers, buds
	or fruit. Collected nematode galls and ?psyllids from this species.
	Collected Aug. 24, 2006. N16.88339 W93.45638, alt. 968 m.

DNA specimens in silica gel

Did not make a list but full details are on each sample bag. Material of *M. calvescens* was collected at all sites located.

Pinned material

Temporary label	Data
WP49-3	<i>Euselasia</i> sp.
	Larva feeding on foliage of Miconia calvescens.
	MEXICO, Chiapas, N16.11685 W91.55996, alt. 985 m
	Collected Aug. 14, 2006, pupated Aug. 16, emerged Aug. 23.
(Additional pinned	material already has permanent labels)

In 95% ethanol

<u>Temporary label</u>	Data
Lacanjá	Psyllids? on shoot tips of Miconia impetiolaris
Chansayab	Near Lacanjá Chansayab, Chiapas (no GPS reading).
	Collected Aug. 18, 2006.
WP48-2	Gregarious lepidoptera larvae on foliage of Miconia calvescens.
WP48-3	Geometrid larva feeding on foliage of Miconia calvescens.
	Collected Aug. 14, 2006, N16.11623 W91.56011, alt. 994 m.
WP49-2	Hairy larva feeding on foliage of Miconia calvescens.
WP58-1	vial broken, specimen lost.
WP58-2	Small adult hemiptera? on foliage of Miconia calvescens.
	Collected Aug. 16 2006, N16.13808 W91.26092, alt. 483 m.
WP62	Psyllids? on foliage of <i>Miconia</i> unidentified sp.
	Collected Aug. 17 2006, N16.11165 W90.92133, alt. 180 m.
WP65	Leaf galls (Cecidomyiidae?) on Miconia impetiolaris
	Near Lacanjá Chansayab, Chiapas (no GPS reading).
	Collected Aug. 18, 2006.
WP69-1	Psyllids? on shoot tips of Miconia calvescens
	Collected Aug. 19, 2006. N17.10310 W91.63666, alt. 577 m.
WP69-1	Hemiptera? eggs on foliage of Miconia calvescens.
	Collected Aug. 19, 2006. N17.10310 W91.63666, alt. 577 m.
WP70	Scale?, "yellow blobs" on foliage of Miconia calvescens.
	Collected Aug. 19, 2006. N17.09490 W91.64741, alt. 644 m.
WP97-1	Nematode galls on <i>Miconia</i> sp. (3 vials)
	Collected Aug. 24, 2006. N16.88339 W93.45638, alt. 968 m.
WP97-2	Psyllids? on shoot tips of <i>Miconia</i> sp.
	Collected Aug. 24, 2006. N16.88339 W93.45638, alt. 968 m.

Live material	
Temporary label	Data The second se
WP48-1	Tachinid puparium emerged from transverse-striped larva feeding on
	foliage of <i>Miconia calvescens</i> . Same species as WP49-3?
	Collected Aug. 14, 2006, N16.11623 W91.56011, alt. 994 m.
WP48-3	Braconid cocoons emerged from large geometrid larva feeding on foliage of <i>Miconia calvescens</i> .
	Collected Aug. 14, 2006, N16.11623 W91.56011, alt. 994 m.
WP49-1	Large hairy larva (<i>Megalopyge</i> ?) feeding on foliage of <i>Miconia calvescens</i> .
	Collected Aug. 14, 2006, N16.11685 W91.55996, 985 m.
WP50-1	Pupa from large green larva (noctuid?) feeding on foliage of Miconia
	calvescens.
	Collected Aug. 14 2006, N16.11162 W91.45116, alt. 838 m.
	Pupated Aug. 23.
WP58	One larva and one pupa similar to WP97-3, feeding on foliage of Miconia
	calvescens.
	Larvae collected Aug. 16, 2006, N16.13808 W91.26092, alt. 483 m.
	One pupated Aug. 25.
WP58-2	Tachinid puparium?
WP62	Larva (geometrid?) feeding on foliage of unident. Miconia sp.
	Collected Aug. 17, 2006, N16.11165 W90.92133, alt. 180 m.
WP97-3	Larvae feeding on foliage of Miconia ?chrysophylla
	Collected Aug. 24 2006, N16.88339 W93.45638, alt. 968 m.

APPENDIX II: Itinerary

- August 10 Flights Edmonton Phoenix Mexico City
- August 11 Visit National Herbarium, Mexico City
- August 12 Flight Mexico City Tuxtla Gutierrez, Chiapas
- Met with Benigno Gómez and Carlos Beutelspacher Rented vehicle and bought supplies for survey
- August 13 By road to San Cristóbal de las Casas
 - and then to Parque Nacional Lagunas de Montebello
- August 14Surveyed in area of Lagunas de Montebello
- August 15 Surveyed along road to Centro Ecoturístico Las Nubes
- August 16 Surveyed along road to Centro Ecoturístico Las Guacamayas
- August 17 Surveyed along road to Lacanjá Chansayab
- August 18 Surveyed in vicinity of Lacanjá Chansayab By road to Chancalá area
- August 19 Surveyed around Metzabok and Nahá natural areas, and by road to Ocosingo
- August 20 Surveyed along road to Palenque
- August 21 Rest day in Palenque
- August 22 Surveyed along road to Tuxtla Gutiérrez
- August 23 Surveyed around areas of El Ocote and Presa Nezahualcoyotl
- August 24 Surveyed around Berriozábal and Laguna Bélgica
- August 25 Packing and preparations for departure
- August 26 Flights Tuxtla Gutiérrez Mexico City Phoenix Edmonton.