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Biological Control of Hamakua Pa-Makani with *Cercosporella* sp. in Hawaii

Eduardo E. Trujillo

University of Hawaii, College of Tropical Agriculture and Human Resources, Department of Plant Pathology, 3190 Maile Way, Honolulu, HI 96822, U.S.A.

Abstract

A Jamaican foliar pathogen of *Ageratina riparia*, incorrectly named *Cercosporella* sp., was introduced into Hawaii on 10 November 1974. Extensive host range terrarium studies under quarantine in 1975 showed this pathogen to be highly host-specific and a safe biological candidate for Hamakua pa-makani control. Permission for its release was granted soon thereafter by the State of Hawaii Board of Agriculture. Host infection occurred at 10–20°C, and > 98% r.h.; 18°C was optimal for infection, disease development and sporulation. Temperatures above 20°C were lethal to the pathogen. The disease is characterized by circular, light chlorotic specks developing into distinct dark to reddish-brown angular lesions with chlorotic margins on the upper leaf surface and massive white sporulation on the lower surface. The pathogen does not grow in normal laboratory media, characteristic of obligate parasites. Inoculum becomes airborne at night when r.h. is >98%; spore distribution is accomplished by wind and rain. Devastating disease epidemics occurred in dense stands of *A. riparia* in cool, high rainfall sites of Oahu, Hawaii, and Maui, 2–3 months after pathogen release. Weed populations were reduced 80% to < 5% in a 9-month period. Similar reduction in weed populations were recorded 3–4 yrs after pathogen release at sites with adequate moisture but temperatures above or below the optimal for disease development. Sites with low temperatures and low rainfall showed > 50% reduction in weed population after 8 yrs of pathogen release. In general, more than 50,000 ha of pasture land have been rehabilitated to their full potential by this pathogen, while no evidence of host resistance nor the presence of mutants pathogenic to other plant species has been encountered in the 8 yrs since its release.

Le Biocontrôle de 'Hamakua Pa-Makani' à Hawaii, au Moyen d'une Espèce de *Cercosporella*

On a introduit à Hawaii, le 10 novembre, 1974, un agent pathogène des feuilles d'*Ageratina riparia*, *Cercosporella* sp., mal identifié d'abord, et originaire de Jamaïque. En 1975, des études poussées de l'éventaibl d'hôtes de cet agent, sous conditions de quarantaine, ont indiqué une haute specificité, de sorte que celui-ci promet d'être un agent sûr pour le contrôle biologique de 'Hamakua Pa-makani'. Peu après, le State of Hawaii Board of Agriculture a autorisé l'élargissement de cet agent pathogène. L'infection des hôtes s'est produite à des temperatures variant de 10° à 20°C, et à plus de 98% r.h., 18°C étant la temperature optimale pour l'infection, le développement de la maladie, et la sporulation. Des temperatures au dessus de 20°C se sont relevées mortelles pour l'agent pathogène concerné. La maladie est caracterisée par des tâches chlorotiques, claires et circulaires, qui se développent en lesions angulaires bien marquées, de couleur foncée ou brun-roux avec des marges chlorotiques, sur le dessus des feuilles et, sur la surface inférieure, une sporulation blanche et massive. L'agent pathogène ne se cultive pas dans les milieux ordinaires de laboratoire, ce qui est caracteristique de tout parasite obligatoire. Les spores se dispersent dans l'air, à l'aide du vent et de la pluie, lorsque l'humidité relative dépasse 98%. Entre 2 et 3 mois après l'élargissement de l'agent pathogène, des colonies d'*A. riparia* de haute densité dans des régions fraiches et très pluvieuses des îles d'Oahu, d'Hawaii et de Maui, ont été ravagés des épidemies de la maladie. Durant une période de neuf mois, les populations de l'herbe nuisible se sont rétrécies de 80% à plus de 95%. On a signalé des réductions semblables, 3 à 4 années après l'élargissement de l'agent pathogène, dans des endroits à humidité suffisante, mais où les temperatures étaient plus elevées ou moins elevées que l'optimum pour le développement de la maladie. Dans des endroits frais et peu pluvieux on a observé < 50% de réductions dans les populations d'herbes après 8 années d'élargissement de l'agent pathogène. En général, plus de 50,000 ha de pâturage ont été completement réhabilités au moyen de cet agent pathogène, au cours des 8 années suivant son élargissement, sans rencontrer ni de résistance de la part des hôtes, ni de presence de mutants nuisibles à d'autres plantes.

Introduction

Ageratina riparia K. & R. (= *Eupatorium riparium* Regel; Compositae), commonly known as Hamakua pa-manaki, was introduced to Hawaii around 1925 (Neal 1965). This member of the Compositae was originally collected from a canyon in El Mirador, Veracruz, Mexico, and taken to Germany by Linden in 1857. From here the plant was spread to other countries as an ornamental (Degener 1946). The species, outside of its range of origin, is a vigorous, hardy plant and prolific seeder; on its native habitat, Hamakua pa-makani could be considered an endangered species (E.E. Trujillo, unpubl. data). Its tiny papose seeds are readily wind disseminated and in moist, cool environments (e.g. forest floor and upcountry grazing lands of the Hawaiian islands), seeds germinate readily and the seedlings, free of natural enemies, grow profusely, dominating in time the natural vegetation covering forest floor and open grazing land (Hosaka *et al.* 1954). In 1973 it was estimated that more than 62,500 ha on the island of Hawaii were infested with this weed (H. Nako, pers. comm.). The infestation was concentrated largely in the Kau-Kona districts from 400–1800 m elevation. In Oahu, the infestation was serious in more than 10,000 ha of the Koolau Range from 300–900 m elevation. The weed also was present on the island of Maui on the Hana highway. In 1973 and 1974 two introduced phytophagous insects, a plume moth *Oidaematophorus* sp. (Lepidoptera: Pterophoridae) and a gall fly *Procecidochares alani* Steyskal (Diptera: Tephritidae), were released by the State of Hawaii Department of Agriculture (DOA) as biological control agents for this plant (Gardner and Davis 1979). The first was collected on *Ageratina pazcuarense* H. & R. (= *Eupatorium pazcuarense* Spregel) in the vicinity of Mexico city and the second on *A. riparia* at finca Mirador, in Veracruz, Mexico (W. Rose, pers. comm.).

A short presentation by the author of the biological control program of *Cassia surattensis* Lamarck (Leguminosae) with *Cephalosporium* sp. (Hyphomycetes) (Trujillo and Obrero 1978) to the Hawaii Cattlemen Council on 10 April 1973, motivated the support of the industry and a grant was received from the Barbara Cox Anthony Foundation to survey Mexico and the Caribbean area for plant pathogens of Hamakua pa-makani. This marked the first organized effort in Hawaii to utilize exotic plant pathogens for the control of weeds. Previously they had been ignored (Templeton and Trujillo 1981).

Methods and Materials

Two land explorations were conducted in Mexico. The first during July to August 1973 covering central Mexico from the U.S. border to Veracruz, and the second during November 1974 covering the Fortin-Jalapa area of Veracruz. The introduction of the pathogen *Cercosporella* sp. (Hyphomycetes) from Jamaica was by permission of Hawaii's Quarantine Section, DOA, and the Federal Animal and Plant Health Inspection Service (APHIS). The original introduction was done by hand-carrying living, diseased host material from Jamaica to Hawaii. Branches of diseased plants were collected before sunrise in the Blue Mountains, placed in a plastic bag, within a double plastic bag containing ice to keep the specimen at low temperature to ensure pathogen survival during the trip. Five leaves with sporulating lesions remained on the branches at arrival in Hawaii 48 h later. These served as the initial inoculum to inoculate four Hamakua and four Maui pa-makani (*A. adenophora* [Spreng.] R. & H.) plants growing in 51 cm (20-inch) diam. terrariums (Lawnware Products, 8220 N. Austin, Morton Grove, IL 60053), kept at a quarantine laboratory at Manoa. Inoculation was done by wetting with a fine brush the underside of the leaves with a spore suspension of the pathogen. Plants in terrariums at 100% r.h. were incubated at 18°C in an air-conditioned room under fluorescent illumination of *c.* 2700 lux.

Inoculum for subsequent studies derived from the original Jamaica inoculum, was maintained on *A. riparia* plants inoculated weekly with a spore suspension containing *c.* 200,000 spores/ml and incubated in terrariums at the environmental conditions previously stated. Inoculum was prepared by washing spores from 14-day-old inoculated leaves into a glass beaker and adjusting to the desired spore concentration with tap water using a spore counting chamber (Howard 1922). Similar inoculum concentrations were utilized for other studies.

All host range studies were conducted in Lawnware No. 340 espherical terrariums (51 cm [20 inches] in diam.). Humidity was adjusted through the manually operated opening at the top. The r.h. was maintained at 100% by adding a liter of water to the floor of the terrarium and closing the top; 98% r.h. was obtained by opening the ventilation window at the top about 2 cm. A battery operated psychrometer manufactured by Environmental Tectonics Corporation was used to measure r.h. Rooted, potted plants of different species tested were inoculated in sets of 4, while 4 Hamakua pa-makani plants inoculated with the same inoculum served as controls. Both leaf surfaces were wetted with tap water spore suspensions of the pathogen using a camel-hair brush. Inoculated plants were incubated at approximately 100% r.h. and 18°C for 24 h, and approximately 98% r.h. and 18°C for 14 days. Fifteen days after inoculation, disease incidence was recorded for inoculated test plants and Hamakua pa-makani controls. Detached *A. riparia* leaves inoculated and incubated at different temperatures in 150 mm diam. disposable petri plates containing a water-soaked paper towel, were used to determine effect of temperature on infection, disease development and sporulation.

Field inoculations were done initially by wetting the leaf surfaces of 100 randomly selected leaves with a camel-hair brush dipped in a spore suspension prepared as previously stated. Sites were selected at different elevations to provide the range of temperatures and humidities necessary for epidemics to occur. Subsequent field inoculations were done by spraying on the host the desired spore suspension with 500 ml plastic spray bottles. Disease was assessed by randomly collecting 10 diseased branches from a 30 m transect of the inoculated area, recording the total number of necrotic diseased leaves over the total number of branch leaves, expressing this as disease percent.

Results and Discussion

Exploration

Mexico's land exploration during July–August 1973 covered more than 10,000 km of roads from the U.S. border to Veracruz. The native plant habitat was located at the end of August in two canyons between Jalapa and Fortin, Veracruz. The specimens found at the time were free of plant pathogens or insect pests. Subsequently the gall fly *P. alani* was collected in this area by William Rose, exploratory entomologist with DOA, and shipped to Honolulu for further studies.

A rust affecting *A. riparia* was discovered in El Mirador, Veracruz, Mexico, in November 1974; however, because of insufficient biological information, this pathogen was not introduced to Hawaii. Explorations conducted in Jamaica during November–December of the same year were successful in finding a previously reported *Cercosporella* sp., the cause of leaf spotting of *A. riparia* in this Caribbean country (Leather 1967). This disease is characterized by circular, light chlorotic specks, to distinct dark reddish-brown, angular lesions with chlorotic margins, on the upper leaf surface (Fig. 1-A). Initial lesions on the lower leaf surface are small, circular to angular, pale green to whitish specks that later become woolly white in appearance, as masses of hyaline *Cercosporella* type nonseptate conidia form (Fig. 1-B). The nonseptate character of the conidia places the pathogen in a genus other than *Cercosporella*. Spores are produced on single hyaline conidiophores arising from hyaline sporodochia. As the spots enlarge, the center becomes necrotic and reddish-brown in color, while sporulation continues on the lesion margin, forming a white ring of spore masses (Fig. 1-C). As the disease progresses, spots become black and coalesce, and leaves necrotize and die. Preliminary pathogen biological information developed during the author's stay at the University of the West Indies, showed fungal characteristics of an obligate parasite. Isolation attempts on common media failed. The pathogen required low temperatures for spore germination on water agar (WA), V8 juice agar (V8) and Difco potato dextrose agar (PDA), where it germinated readily at 10, 14, 16 and 18°C. Although germ tube development occurred at these temperatures, no mycelium was produced or colonies developed. Spore germination ceased at temperatures higher than 20°C.

Field data gathered during exploration of the Blue Mountains and the northeastern coast of Jamaica pointed to a high pathogen specificity, none of the major crops examined nor representative species of more than 20 families of plants growing adjacent to diseased *A. riparia* showed evidence of white spore masses on the lower leaf surface, characteristic signs of the pathogen. As a result, the original introduction of the pathogen from Jamaica to Hawaii was done in living plant material.

Host Range Studies

The first visible symptoms on Hamakua pa-makani plants inoculated with the original Jamaica inoculum and incubated in terrariums at 18°C under incident illumination > 2700 lux appeared 9 days after inoculation. These showed as small, chlorotic specks on the upper leaf surface, becoming necrotic 12–13 days after inoculation. On the lower leaf surface the first visible signs of the pathogen appeared 7 days after inoculation as pinpoint, whitish protuberances immediately below the stomata. Microscopic examination showed these to be hyaline sporodochia filling the substomatal chamber. Subsequently, sporulation occurred abundantly, and by the 11th day the lesions on the underside of the leaves were covered with the characteristic white spore masses. No lesions developed on *A. adenophora*-inoculated plants; repeated pathogenicity tests gave similar negative results.

A. riparia was the only host found for *Cercosporella* sp. Inoculation tests on 40 plant species representing 29 families were all negative, while inoculated *A. riparia* controls always were positive. Inoculations involved: ti, *Cordyline terminalis* (L.) Kunth (Agavaceae); mango, *Mangifera indica* L. (Anacardiaceae); plumeria, *Plumeria obtusa* L. (Apocynaceae); anthurium, *Anthurium andraenum* Lind. (Araceae); pineapple, *Ananas comosus* (L.) Merr. (Bromeliaceae); papaya, *Carica papaya* L. (Caricaceae); beet, *Beta vulgaris* L. (Chenopodiaceae); calendula, *Calendula* sp., mums, *Chrysanthemun* sp., Maui pa-makani, *A. adenophora*, lettuce, *Lactuca sativa* L., ligularia, *Ligularia*

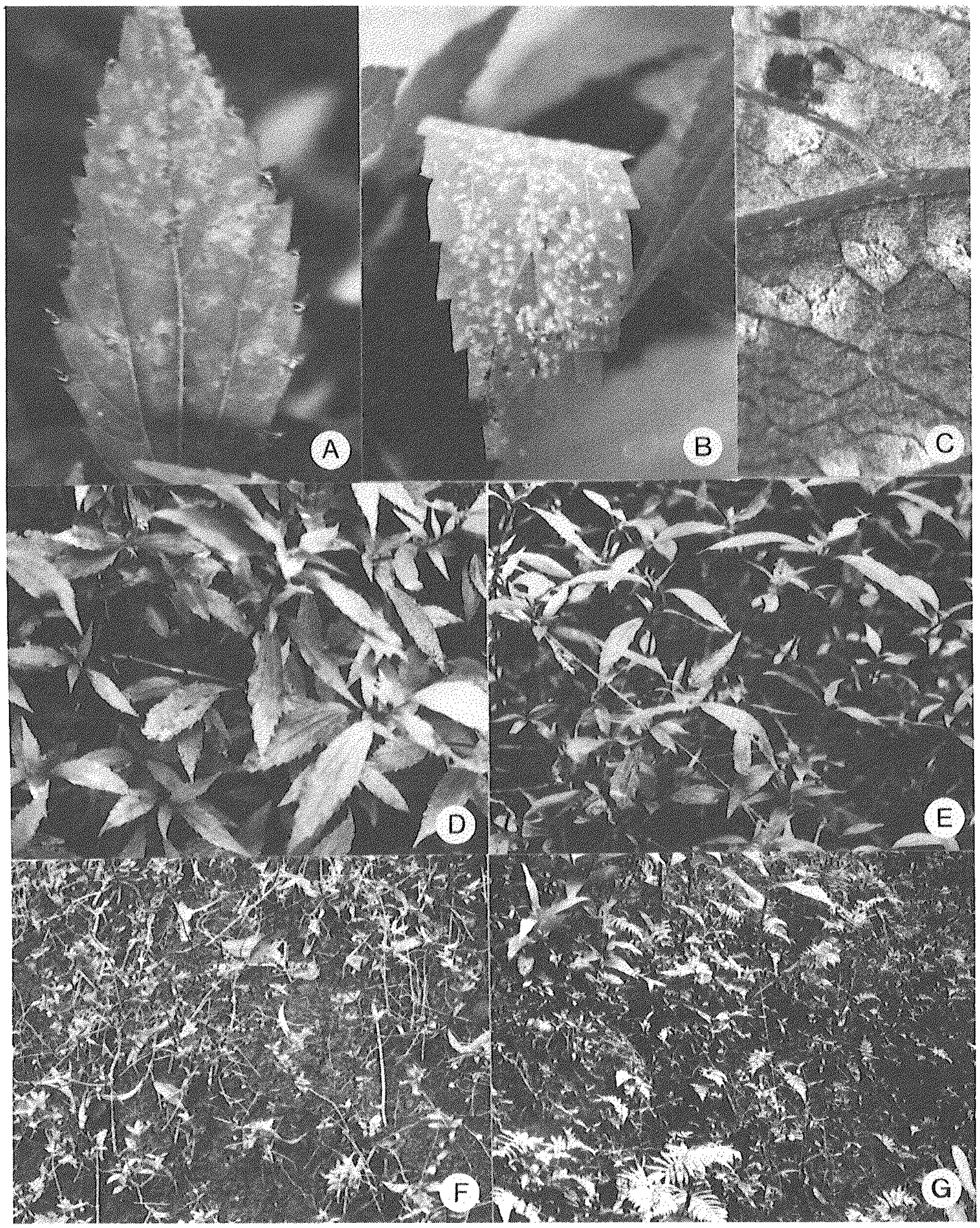

Fig. 1. '*Cercosporella*' blight of Hamakua pa-makani. (A) *Ageratina riparia* K. & R. upper leaf surface showing initial and advance symptoms of the disease. (B) Lower leaf surface showing white spore masses, sign of the pathogen. (C) Lower leaf surface showing initial and advanced necrotic spots magnified 8×. (D–G) Effects of disease epidemics on weed at Tantalus, Oahu, 1975–76, where ideal conditions for these prevailed: (D) 6 wks; (E) 10 wks; (F) 4 months; and (G) 6 months after pathogen release.

tussilaginea (Burm. f.) Makino, marigold, *Tagetes* sp., wedelia, *Wedelia trilobata* (L.) Hitchc. (Compositae); sweet potato, *Ipomoea batatas* (L.) Lam., morning glory, *Ipomoea* sp. (Convolvulaceae); head cabbage, *Brassica oleracea capitata* L. (Cruciferae); cucumber, *Cucumis sativus* L. (Curcurbitaceae); poinsettia, *Eurphorbia pulcherrima* Willd. (Euphorbiaceae); bamboo, *Bambusa* sp., sugar cane, *Saccharum officinarum* L. (Varieties 37–1933, 54–775 and Hawaiian purple), maize, *Zea mays* L. (Graminae); coleus, *Coleus blumei* Benth. (Labiatae); avocado, *Persea americana* Mill. (Lauraceae); beans, *Phaseolus vulgaris* L. (Leguminosae); hibiscus, *Hibiscus* sp. (Malvaceae); banana, *Musa* sp. (Musaceae); ohia lehua, *Metrosideros collina* (Fort.) Gray subsp. *polymorpha* (Gaud.) Rock., guava, *Psidium guajava* L. (Myrtaceae); dendrobium, *Dendrobium* sp. (Orchidaceae); coconut, *Cocos nucifera* L. (Palmaceae); lilikoi, *Passiflora edulis* Sims (Passifloraceae); phlox, *Phlox* sp. (Polemoniaceae); macadamia, *Macadamia ternifolia* F. Muell. (Proteaceae); rose, *Rosa* sp. (Rosaceae); coffee, *Coffea arabica* L., gardenia, *Gardenia jasminoides* Ellis (Rubiaceae); lime, *Citrus* sp. (Rutaceae); litchi, *Litchi chinensis* Sonn. (Spindaceae); snap dragon, *Antirrhinum* sp., (Scrophulariaceae); tomato, *Lycopersicon esculentum* Mill., egg plant, *Solanum melongena* L. (Solanaceae); lantana, *Lantana camara* L., jamaica vervain, *Stachytarpheta jamaicensis* (L.) Vahl. (Verbenaceae).

Optimal Temperature for Epidemics

Inoculation studies conducted on detached leaves in moist chambers at 100% r.h. and 10, 12, 14, 16, 18, 20 and 22°C showed 18°C as the optimal temperature for spore germination, infection, sporodochial formation, sporulation and symptom development. Spore germination and infection occurred from 10–20°C; no spore germination or infection occurred at temperatures higher than 20°C. Inoculation of detached pa-makani leaves by wetting the lower surface with *c.* 0.1 ml suspension containing *c.* 20,000 spores and incubated at 10, 12, 14, 16, 18, 20 and 22°C, and 100% r.h. for 20 days showed an average of 4, 13, 120, 220, 652, 198 and 0 lesions/leaves. At 18°C sporodochia was visible in 7 days and abundant sporulation occurred in 10–11 days after inoculation. At 10 and 12°C, sporodochial development was very slow, becoming visible with whitish protuberances on the leaf lower surface after 20 and 16 days, and white spore masses became visible in 30 and 20 days after inoculation, respectively. At incubation temperatures of 14, 16, 18 and 20°C, sporodochia formed and sporulation was observed 11, 9, 7 and 9 days after inoculation, and white spore masses became visible at 13, 12, 8 and 10 days, respectively. No infection occurred at 22°C as spores on the surface of the leaves failed to germinate. Microscopic observation of Acid fuchsin stained epidermal strips from inoculated leaves showed only ungerminated spores.

Spores subjected to temperatures higher than 24°C lost viability rapidly as temperature increased. Nine sets of three detached diseased leaves exhibiting more than 200 sporulating lesions were incubated at temperatures of 24, 28 and 32°C for 1, 5 and 24 h periods. Spores were removed from the lesions with a wet camel-hair brush and inoculated on nine sets of three healthy leaves and incubated for 18°C and 100% r.h. for 10 days. Spores survived 5 h at 24°C without noticeable loss in viability, as shown by the equal number of lesions formed on inoculated leaves by spores treated for 1 and 5 h to this temperature, while a 50% drop in viability was recorded for spores treated for 24 h; there was a 50% drop in the number of lesions produced by spores subjected to this treatment. Spores survived temperatures of 28–32°C for 1 h without appreciable loss in viability. Longer treatment times reduced viability significantly.

Effect of temperature on pathogen sporulation on host tissues was measured on detached diseased Hamakua pa-makani leaves. These were thoroughly washed with tap water and a camel-hair brush to remove spores present and dried with absorbent paper before incubating for 24 h at 14, 16, 18, 20 and 22°C and 100% r.h. Sporulation was abundant at 18, reduced by about one half at 16 and 20, hardly detectable at 14, and absent at 24°C. Attempts to determine the inoculum potential necessary to cause infection were made by inoculating a known spore concentration to the undersurface of 20 detached leaves incubated at 16°C. Lesions were counted 15 days after inoculation. The results of this experiment showed an inoculum potential of 70 spores/lesion.

Field Release and Biological Control Potential

In October 1975, the Hawaii Board of Agriculture, DOA, granted permission for field release of the *Cercosporella* sp. introduced from Jamaica as a biological control agent for Hamakua pa-makani. On 4 November 1973, plants were inoculated at three sites on the Tantalus Ridge of Oahu, at elevations of 450, 500 and 550 m. An initial inventory of the pa-makani population showed these sites to be 80% covered with the weed. In each site the leaves of 100 plants were wetted with a spore suspension containing *c.* 200,000 spores/ml. Lesions were visible 12 days after inoculation at 450 and 500 m elevation, and at 550 m 11 days after inoculation. Evidence of secondary infections were recorded at all sites 20 days after inoculation by the appearance of new lesions on a few non-inoculated leaves. Twenty-day-old lesions at all sites were about 2 mm in diam. and could be recognized by the necrotic centers already present, while secondary lesions showed as very small white specks. Sporulation was sparse at 450 m where temperatures were too high for the pathogen, while it was abundant at 500–550 m elevations with favorable temperatures.

No evidence of severe disease epidemic was recorded during the first 6 wks after inoculation in a 100 m radius around the inoculated site. However, after exploring the 9 km trail that goes from the western side of the Tantalus road to Pauoa flats and the ridge summit at 670 m elevation, evidence of extensive disease spread was recorded. During the first 300 m of trail no evidence of disease spread was noticed. The first evidence of disease was recorded about 550 m away from the inoculation site, where less than 1% of the foliage had small necrotic lesions; sporulation was massive on the underside of the lesion and some evidence of secondary infection was evident in some leaves. Approximately 1.6 km from the inoculation site and 600 m elevation, the first area with more than 5% diseased leaves was encountered. The greatest disease incidence was found at 650 m elevation, 4 km from the initial inoculation sites, where more than 10% of the leaves showed necrotic lesions. The rainfall at this elevation averages 7.112 m annually with temperature of 16°C at night, ideal conditions for epidemics of the disease to develop. A day after this finding, the St. Louis ridge, located 5 km to the northeast of Tantalus was surveyed. The disease was recorded at about 550 m elevation rather extensively distributed among the weed population of this uninoculated area. Subsequently, airborne spores of the pathogen were trapped on Rotorod air samplers (manufactured by Ted Brown Associates, 26338 Esperanza Dr., Los Altos Hills, CA 94022), used on Tantalus at 1.5 m from the ground from 7–7:30 pm when r.h. was $> 95\%$. No spores were trapped in air sampled from 8:00 a.m. to 6:30 p.m., when r.h. was $< 95\%$. The rapid spread of the pathogen in the area was possibly due to the normal air movement in Tantalus, typically downdraft in the evening and updraft early in the morning. A tropical storm which affected Oahu during 22–27 November 1975, with high winds and more than 300 mm of rain, provided optimal conditions for pathogen distribution and disease. The minimal-maximal temperatures for Tantalus during December were 12–24°C, ideal for infection and sporulation.

Monthly assessments of weed population at the initial Hamakua pa-makani inoculation sites at Tantalus showed a rapid decrease in foliage and plant vigor at all sites and the weed population dropped from 70–80% to < 1% in 9 months. In August 1976, most of the space once occupied by the weed was covered in other plant species such as ferns, grasses and indigenous plants (Fig. 1, D–G).

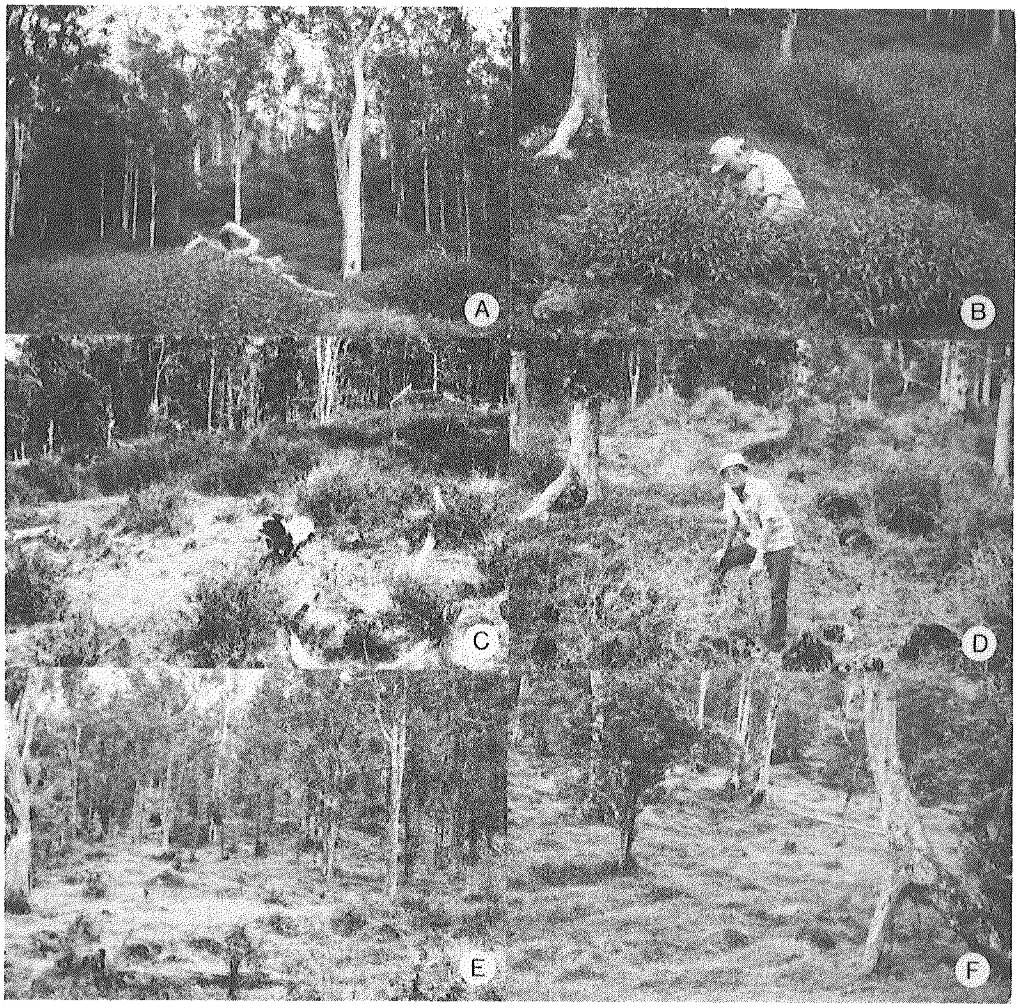

Fig. 2. Palani ranch, Kona, island of Hawaii, inoculation site at 900 m elevation, where low precipitation prevails during summer months. (A, B) Degree of Hamakua pa-makani infestation 10 November 1975, previous to pathogen release. (C) Weed population November 1976, showing recovery after first disease epidemics. (D) Degree of weed control, August 1979, 3 yrs after pathogen release. (E, F) 99% Hamakua pa-makani control January 1983, approximately 8 yrs after pathogen release.

On 10 November 1985, a water suspension of spores at 16°C was carried by plane to Hawaii and was used to inoculate by wetting the surface of 100 leaves at 4 sites in the upcountry range lands of Palani ranch, Kona. Approximately 500 ml of inoculum containing 200,000 spores/ml were used at 900, 1100, 1350, and 1500 m elevation. The areas selected for this study were 70–80% invaded by Hamakua pa-makani (Figs. 2-A, B; 3-A, D). Observations made 15 days later showed well developed lesions at inoculation sites 900 and 1100 m elevation, but none at 1350 and 1500 m. Lesions were visible at the higher elevations 30 days after inoculation and instead of chlorotic specks on the upper leaf surface the symptoms appeared as purple spots. The anthocyanin formation around the lesions was attributed to prevailing low temperatures (< 10°C at night).

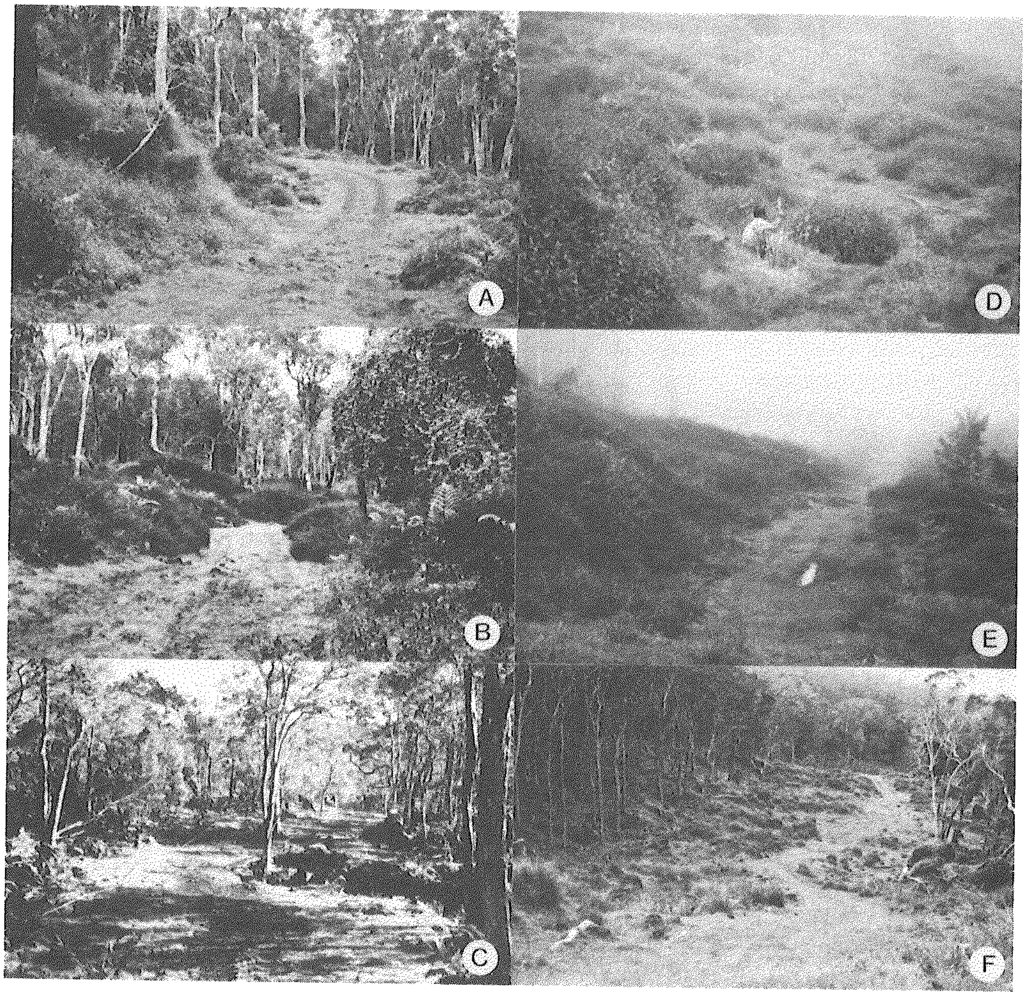

Fig. 3. Palani ranch, Kona, island of Hawaii, inoculation sites with high rainfall during the summer months, at 1000 m (A–C) and 1500 m (D–F) elevations. (A, D) Weed infestation previous to pathogen release 10 November 1975. (B, E) Massive disease epidemics May 1976, approximately 7 months after pathogen release. (C, F) Degree of Hamakua pa-makani control September 1978, approximately 3 yrs after pathogen release.

On 26 February 1976, a water suspension of > 200,000,000 spores was sprayed at random on *A. riparia* plant population within 500 ha of Palani ranch land, ranging in elevation from 600–1800 m. Weed population was estimated at 70–80% of the total biomass of the area. Monthly disease surveys showed 5, 20, 50 and 95% disease, respectively, 2, 3, 4 and 6 months after inoculation. The epidemic of the disease was so severe that the plants had very sparse foliage by the end of November and the following year blooming was noticeably reduced. The disease spread to adjacent non-inoculated weed areas by means of airborne spores which were blown in all directions by prevailing wind currents and deposited later by rain on the host. Four months after the initial inoculations in the island of Hawaii the pathogen was found on Hamakua pa-makani plants growing on Maui, in the vicinity of the Seven Sacred Pools, some 120 km from Kona, across the Hawaii-Maui Channel.

The disease initially developed more rapidly at the 900 and 1100 m sites than at 1350 and 1500 m; temperatures were more favorable at the lower elevations during the winter months. As summer temperatures began to climb, the disease incidence increased at the higher elevation and control of the plant became noticeable in the humid high country. The control of Hamakua pa-makani by *Cercosporella* sp. at Palani ranch has been extremely successful, and where there was 70–80% pa-makani there is now 70–80% Kikuyu grass pasture (Figs. 2–3).

Annual disease surveys done in Hawaii have shown > 95% control of the weed in less than a year after inoculation, on most areas with average annual rainfall of > 1.8 m and optimal temperatures for disease development during the wet periods. In areas with adequate moisture but lower than optimal temperatures for disease development, control > 95% was attained 3–4 yrs after inoculation. In areas with lower or higher than optimal temperatures for disease and low rainfall, the control has been < 80% after 8 yrs of pathogen release.

The successful biological control of *A. riparia* with *Cercosporella* sp. in Hawaii correlates with a number of favorable epidemiological factors influencing the rate of inoculum increase and distribution; e.g. high rainfall at the initial release sites; prevalence of temperatures favorable to the pathogen; and presence of a large infestation of Hamakua pa-makani. The rapid distribution of the pathogen soon after release was similar to the distribution of the skeleton weed rust in the Wagga Wagga area of Australia (Cullen *et al.* 1973). However, it was accomplished in less time and perhaps with fewer generations of the pathogen. The devastating ensuing epidemics in wet, optimal temperature sites of Oahu, Hawaii and Maui compared favorably with the late blight epidemics that destroyed Ireland's potato crops in 1945–46 (Large 1946). These results have led to the conclusion that host-specific fungal pathogens are safe and beneficial tools in exotic weed control. This is supported by the evidence that after 8 yrs of pathogen release the fungus still remains host-specific under many environmental conditions.

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