The Resistance of Mango (*Mangifera indica*) Cultivars to Tip Dieback Disease in Florida

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ABSTRACT

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The resistance of *Mangifera indica* to tip dieback disease caused by *Botryosphaeria ribis*, anamorphic state *Fusicoccum* sp., was determined on 361 trees of 122 mango cultivars and relatives planted in the mango germ plasm collection at the University of Florida, Tropical Research and Education Center, Homestead. Three trees from each cultivar were evaluated on a 1 to 5 scale ranging from those free of tip dieback to those with extensive branch necrosis. Each cultivar was assigned to one of eight major systematic groups based on race or geographic origin. Resistance to tip dieback disease was not associated with any of the groups. No significant differences in mean disease severity were found among the Indian, Southeast Asian, West Indian, Haden, or Sandersha parts of the Haden-Sandersha Complex, Turpentine types, or a group of unclassified cultivars. Two *Mangifera* species (*M. odorata* and *M. zeylanica*) showed the lowest mean disease rating. Cultivars were also grouped into monoembryonic and polyembryonic types. Although several mango cultivars, mainly found in the monoembryonic group, appeared to be free of the disease, resistance to tip dieback was not associated with embryony. This study provides the first indication that field resistance to tip dieback may be present in some mango cultivars.

Additional keywords: adventive and zygotic embryos, Ascomycotina, *Botryosphaeria dothidea*, genetics

Tip dieback of mango (*Mangifera indica* L.), frequently included under the broad term mango decline (11,12,13,18), is a prevalent disease that has been affecting mango trees in southern Florida since the late 1970s. The etiology of mango dieback remained unknown for several years. McSorley et al. (11,12) consistently found high populations of the nematode *Hemicriconemoides mangiferae* Siddigi in soils around old, declining mango trees. The association between *H. mangiferae* and tree condition was, however, not evident in young trees, and declining trees were observed in young groves where very few *H. mangiferae* were present, suggesting that decline of young trees is due to other factors (12). The association of host nutrition with decline was investigated by Schaffer (18,19). He concluded that manganese and iron were limiting elements in affected trees, but the authors (13) did not determine whether the deficiency of these microelements actually caused mango decline.

The pathological nature of tip dieback was revealed for the first time during

pathogenicity studies with fungi isolated from symptomatic mango tissues (15). The primary causal organism of the disease was identified and described as *Botryosphaeria ribis* Grossenbacher & Duggar anamorphic state (6). The identity of *B. ribis* was confirmed by B. C. Sutton, International Mycological Institute (IMI), Kew, Surrey, England, with a herbarium specimen deposited as IMI 293744. No name has been formally assigned to the anamorph, but according to the current taxonomic concepts, *Fusicoccum* sp. is the correct generic name for it (E. Punithalingam, International Mycological Institute, *personal communication*). *B. ribis* induced severe dieback in up to 83% of the inoculated plants (15). Several other fungi were occasionally isolated from symptomatic tissues of mango showing dieback symptoms, including *Alternaria* sp., *Colletotrichum* sp., *Curvularia* sp., *Fusarium equiseti* (Corda) Sacc., *Macrophoma* sp., *Oidium* sp., and *Stigmata* sp. Moreover, *Pestalotiopsis* sp. and *Chalaropsis* sp. were isolated from walls of ambrosia beetle larvae bark galleries found in mango trees. In the pathogenicity studies, some of these fungi caused necrosis around inoculation scars, but none of them induced dieback. The avenues of penetration and infection of unwounded stems or twigs by *B. ribis* are unknown (15). Nevertheless, it seems probable that the host can be weakened by these secondary pathogens that cause external lesions and

can facilitate the entrance and infection of endophytic fungi. In addition, because symptoms of mango tip dieback develop after long incubation periods, generally 5 to 10 months (15), infection and disease development are often related to environmental stresses suffered during the incubation period (L. J. Ramos and S. P. Lara, *unpublished*). *Diplodia* sp., another anamorphic state of *Botryosphaeria*, induced mild dieback symptoms in up to 17% of inoculated plants (15). *Diplodia* sp. has been previously reported in Florida mangoes (21).

B. ribis appears to be an aggressive pathogen that invades the vascular system of mango plants. Histopathological studies of symptomatic tissue revealed the production of tyloses in xylem vessels, necrosis of xylem vessels, formation of dark inclusions, and the presence of fungal hyphae (15). These findings suggested that *B. ribis* infection brings about a disjunction of xylem elements with concomitant restriction of water and mineral transport. *B. ribis* was associated with dieback of young stems and limbs in the Hawaiian Islands as early as 1929 (22).

After artificial inoculations of Keitt mango with different fungal isolates, Ploetz et al. (13) established the relationships of several fungi with all or several mango decline symptoms, such as bud necrosis, tip dieback, gummosis, and vascular discoloration. The authors reported a relatively low incidence (10%) of tip dieback after inoculations with *Dothiorella dominicana* Petr. & Cif. (synonym: *Fusicoccum aesculi* Corda; teleomorph: *Botryosphaeria dothidea* (Moug:Fr.) Ces & De Not.) and 12% incidence after inoculations of *Lasiodiplodia theobromae* (Pat.) Grifford & Maubl. (synonym: *Diplodia natalensis* Pole-Evans, *Diplodia theobromae* Pat.; teleomorph: *Botryosphaeria rhodina* (Cooke) Arx). An even lower incidence (4%) of tip dieback was obtained after inoculations with *Colletotrichum gloeosporioides* (Penz.) Penz. & Sacc. and *Alternaria alternata* (Fr:Fr.) Keissl.

Ramos et al. (15) determined *B. ribis* (anamorph: *Fusicoccum* sp.) and, less commonly, *Diplodia* sp. as the primary causal organisms of tip dieback; whereas Ploetz et al. (13) reported *B. dothidea* (anamorph: *Fusicoccum aesculi*) and *Diplodia theobromae* as the fungi most commonly causing tip tieback. B. C. Sutton suggested that *Fusicoccum aesculi*, the

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Table 1. Severity of tip dieback of mango (*Mangifera indica*) cultivars and relatives planted in the collection at the University of Florida, Tropical Research and Education Center, Homestead

Cultivar	Embryo type ^a	Disease rating ^b	Cultivar	Embryo type	Disease rating
1. Indian types					
Ameeri	M	2.33 ± 0.22	Langra Benarsi	M	1.00 ± 0.00
Amini	M	3.33 ± 0.22	Mulgoba	M	3.00 ± 0.00
Bennett	M	1.67 ± 0.44	Paheri	M	1.67 ± 0.44
Bombay Green	M	2.00 ± 0.22	Rajpuri	M	1.33 ± 0.22
Borsha	M	1.33 ± 0.22	Safeda Lucknow	M	3.00 ± 0.00
Gola	M	1.00 ± 0.00	Sandersha	M	1.00 ± 0.00
Joe Welch	M	2.67 ± 0.22	White Langra	M	1.00 ± 0.00
2. Southeast Asia types					
Aroemanis CR-1	P	1.67 ± 0.22	Madoe CR-42	P	2.33 ± 0.22
Cambodiana	P	3.00 ± 0.38	Micongensis	P	2.33 ± 0.22
Cecil	P	2.00 ± 0.38	Mun	P	1.33 ± 0.22
Duncan	M	2.00 ± 0.38	Nam-Dok-Mai	P	2.67 ± 0.38
Florigon	P	3.33 ± 0.22	Okrun	P	3.00 ± 0.38
Gadoeng	P	2.67 ± 0.79	Philippine	P	2.33 ± 0.22
Golek-CR-1	P	1.33 ± 0.22	Rockdale Saigon	P	1.00 ± 0.00
Hindi	P	2.67 ± 0.22	RosiGold (M13264)	M	1.67 ± 0.00
Honc Cambodiana	P	3.00 ± 0.00	Tutehau (PI 99804)	P	2.67 ± 0.58
James Saigon	P	1.33 ± 0.22			
3. West Indian types					
Brander	P	3.00 ± 0.22	Mammee	P	3.00 ± 0.22
Julie	M	3.00 ± 0.00	Manzano	P	1.33 ± 0.22
Kurashige	P	1.00 ± 0.00	Sophie Fry	M	1.33 ± 0.22
Madame Francis	P	3.67 ± 0.22			
4. Haden-Sandersha Complex—Haden part					
4 Way Bombay	M	1.67 ± 0.22	Hodson	M	2.67 ± 0.22
Adams	M	2.33 ± 0.22	Irwin	M	2.67 ± 0.22
Jacquelin	M	2.67 ± 0.38	Irwin sdg # 2	M	2.67 ± 0.22
AG Reasoner	M	1.33 ± 0.22	Joellen	P	1.00 ± 0.00
Alice	M	1.00 ± 0.00	Kent	M	2.00 ± 0.38
Bill Herman	M	2.33 ± 0.22	Lathrop	M	1.00 ± 0.00
Burgner	M	3.00 ± 0.00	Lippens	M	3.00 ± 0.00
Carrie	M	2.33 ± 0.22	Lucille	M	3.33 ± 0.58
Cogshall	M	1.00 ± 0.00	Miami Late	M	2.67 ± 0.22
Earle No. 1	M	1.00 ± 0.00	Momi-K	M	3.33 ± 0.22
Edward	M	1.33 ± 0.22	Monser	M	2.00 ± 0.00
Ed × Earle-15	P	2.33 ± 0.22	Ono	P	1.33 ± 0.22
Ed. × Kent 14	M	1.67 ± 0.22	Pettigrew	M	1.00 ± 0.00
Eldon	M	1.33 ± 0.22	Pillsbury	M	4.00 ± 0.00
Fascell	M	1.33 ± 0.22	Pillsbury A	M	1.67 ± 0.22
Glenn	M	2.67 ± 0.58	Pope	M	3.00 ± 0.00
Glazier	M	3.33 ± 0.22	Pruter	M	1.00 ± 0.00
Gouvea	M	3.00 ± 0.00	Reasoner	P	2.00 ± 0.38
Graham	M	2.67 ± 0.22	Simmonds	P	1.33 ± 0.22
Haden	M	1.67 ± 0.44	Sunset	M	2.33 ± 0.22
Hansen	M	1.00 ± 0.00	Vandyk	M	2.33 ± 0.22
Harris	M	2.00 ± 0.00	Wallace	M	2.67 ± 0.22
Harris sdg	M	1.67 ± 0.22	Zill	M	2.33 ± 0.20
5. Haden-Sandersha Complex—Sandersha part					
Anderson	M	2.33 ± 0.22	Parris	M	1.00 ± 0.00
Beverly	M	2.00 ± 0.00	Parvin	M	2.00 ± 0.00
Brooks	M	2.33 ± 0.22	Ruby	M	1.00 ± 0.00
Davis Haden	M	2.00 ± 0.00	Rycheck	M	2.33 ± 0.44
Dixon	M	2.67 ± 0.79	Sandersha sdg 6-16	M	1.33 ± 0.22
Early Gold	M	2.67 ± 0.22	Scuppernong	M	3.33 ± 0.22
Ford	M	2.00 ± 0.00	Sensation	M	2.33 ± 0.22
Golden Brooks	M	2.67 ± 0.22	Smith	M	1.67 ± 0.44
Keenan	M	1.00 ± 0.00	Springfels	M	2.33 ± 0.22
Keitt	M	2.00 ± 0.00	Valencia Pride	M	1.33 ± 0.22
Martin	M	1.00 ± 0.00	Webber	M	1.33 ± 0.22
Osteen	M	4.00 ± 0.44			
6. Turpentine types					
Itamaraca	P	3.00 ± 0.38	Saber	P	2.50 ± 0.22
No. 11	P	3.00 ± 0.65	Stringless Peach	P	2.67 ± 0.22
Peach	P	2.00 ± 0.38	Turpentine	P	4.00 ± 0.38
Red Itamaraca	P	2.33 ± 0.27			
7. Nonclassified cultivars					
Golden Delight	M	2.67 ± 0.22	Ott	M	2.67 ± 0.58
Kensington	P	1.33 ± 0.22	Wally	M	1.67 ± 0.22
8. Other <i>Mangifera</i> species					
<i>M. odorata</i>	M	1.67 ± 0.22	<i>M. zeylanica</i>	M	1.67 ± 0.44

^a Embryo types (M = monoembryonic and P = Polyembryonic) were randomly distributed.

^b Mean disease ratings followed by standard errors (ratings 1 to 5: 1 = free of tip dieback; 5 = severely damaged plants). No significant differences among eight major race, groups of complexes, GLM, nested design, using Type III MS cultivar (group) as an error term. (df = 7, *F* value = 1.10, *P* > 0.369). There were significant differences in disease response among individual cultivars using MSE as the error term (df = 121, *F* value = 3.52, *P* > 0.0001). The three replicated trees from each cultivar were not distributed at random. Major groups were randomly distributed.

anamorphic state of *B. dothidea* causing canker and dieback in *Tetragastris panamensis* (Engl.) Kuntze, is morphologically different from the anamorphic state of *B. ribis*, which some authors consider to be a synonym of *B. dothidea* (4). There is disagreement in the literature regarding the delimitation of both the genus and the species within the genus *Botryosphaeria* (7). Discrepancy in the species identified by Ramos et al. (15) and Ploetz et al. (13) can be explained by their use of different sources for taxonomic identification and confirmation of isolates. The isolates of Ramos et al. (15) were identified and confirmed by B. C. Sutton; whereas the isolates of Ploetz et al. (13) were identified and confirmed by G. I. Johnson (Queensland Department of Plant Industries, Indooroopilly, Australia).

The Tropical Research and Education Center (TREC), University of Florida, is a repository for numerous monembryonic and polyembryonic mango cultivars representing diverse genetic backgrounds from the world's major races and complexes of mango and its relatives. (14,16). Rhodes et al. (16) constructed phenograms, based on cluster analysis, to assign cultivars to specific systematic groups. Grouping was based on 73 vegetative and reproductive characters, with emphasis on fruit, inflorescence, leaf morphology, and embryony. Combined with evidence of the pathological nature of tip dieback, the trees in the collection at TREC provided the opportunity to investigate the possibility that resistance to tip dieback disease was present in some of these systematic groups. The objective of this study, therefore, was to investigate possible sources of field resistance to tip dieback in individual mango cultivars and to determine if field resistance is associated with a systematic group or embryony of the cultivars.

MATERIALS AND METHODS

Plant material. The mango germ plasm collection at TREC is a repository for 122 cultivars. The trees are approximately 20 years old and are planted in rows spaced 7.6 m apart in Rockdale soil (Krome very gravelly loam soil). All cultivars were grafted on Turpentine, polyembryonic nucellar rootstocks. To determine if resistance to tip dieback disease was associated with any major systematic group, each cultivar was assigned to one of the following groups according to phenotypic characteristics previously described by Popenoe (14) and Rhodes et al. (16): (1) Indian types with monoembryonic, roundish fruit; (2) Southeast Asia types, mainly from Indochina, Java, Pacific Island, Philippines, South Vietnam, and Thailand, with polyembryonic, oblong, elongated fruit; (3) West Indian types, mainly from Barbados, Jamaica, and Puerto Rico, with intermediate fruit shapes; (4) Haden-Sandersha Complex—Haden part, hybrids from Flor-

ida and Hawaii with large elliptic-shaped fruit; (5) Haden-Sandersha Complex—Sandersha part, hybrids of Florida and Hawaii with large oblong-shaped fruit; (6) Turpentine types, polyembryonics, mainly from Cuba, with small, elongated fruit; (7) nonclassified cultivars; and (8) other *Mangifera* species (*M. odorata* Griff. from Southeast Asia and *M. zeylanica* Hook from Ceylon). The subdivision of the Haden-Sandersha Complex described by Rhodes et al. (16) into Haden part and Sandersha part is somewhat artificial. It was based mainly on Popenoe's (14) division of mangos grown in Florida into three types: Mulgoba, Alphonse, and Sandersha.

The cultivars were also grouped in the present study as either monoembryonic or polyembryonic according to their embryo type to study the influence of embryony on

resistance to tip dieback (Table 1). Classification of cultivars by embryonic type was based on published (1,16) and nonpublished (C. W. Campbell, R. J. Knight, and R. J. Schnell, *personal communications*) data.

Trees were fertilized with 8-3-9 (NPK) three times per year. Iron chelate (Geigy Sequestrene 138) and minor element sprays were applied periodically to soil and foliage, respectively. Benomyl, chlorothalonil, copper fungicides, and high-volume overhead irrigation were periodically applied to the trees.

Disease evaluation. The presence or absence of tip dieback was recorded for 361 trees in the germ plasm collection representing three replicates of most cultivars. The severity of dieback symptoms in twigs and branches on individual trees was rated

using a scale from 1 to 5, where 1 = trees free of disease; 2 = an early stage of infection characterized by browning of leaf petioles and midveins and presence of distal or marginal leaf blade necrosis in one or two branches; 3 = the presence of dead leaves, which may remain attached, in the tips of several branches, vascular browning, and evidence of pathogen invasion of vascular tissues, formation of tyloses in xylem vessels, and dark inclusions and fungal hyphae present in stem sections; 4 = dead leaves and progressive defoliation extending to many larger branches along with increased severity and spread of vascular symptoms; and 5 = severe dieback that extended to major portions of the tree. The trees were evaluated for progression of disease symptoms in December 1988 to 1990. Because of the progressive nature of

Rows	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	2 RosiGld	1 RosiGld	2 RosiGld	2 Wally	2 Wally	2 Wally	2 Vandyke	2 Vandyke	3 Vandyke	*	*	*	*	*	3 Irwin 2	2 Irwin 2
2	1 Red Itam	2 Red Itam	*	2 M.Odor.	2 M.Odor.	1 M.Odor.	1 M.Zey.	3 M.Zey.	1 M.Zey.	2 Philip.	3 Philip.	2 Philip.	4 Ott	1 Ott	3 Ott	3 Irwin 2
3	1 No.11	4 No.11	4 No.11	2 S. Peach	4 S. Peach	2 S. Peach	1 Peach	3 Peach	2 Peach	2 Saber	3 Saber	*	3 Itam.	2 Itam.	4 Itam.	*
4	3 Mamm.	*	3 Mamm.	3 Brander	3 Brander	2 Brander	1 Ono	1 Ono	1 Ono	1 Kurash.	1 Kurash.	1 Kurash.	3 Julie	3 Julie	4 Julie	4 Turp.
5	3 Mamm.	1 Kensig.	2 Kensig.	1 Kensig.	1 Parris	1 Parris	1 Parris	2 S. Fry	1 S. Fry	1 S. Fry	1 Manzano	1 Manzano	2 Manzano	3 Tuehau	4 Tuehau	1 Tuehau
6	3 S.Luck.	1 Sander.	1 Sander.	1 Sander.	1 L.Benars.	1 L.Benars.	1 L.Benars.	1 Gola	1 Gola	1 Gola	3 Joe W.	3 Joe W.	2 Joe W.	4 M.Fran.	4 M.Fran.	3 M.Fran.
7	*	3 S.Luck.	4 Amini	3 Amini	3 Amini	1 Borsha	1 Borsha	2 Borsha	3 Borsha	3 Mulgoba	3 Mulgoba	3 Mulgoba	1 Paheri	1 Paheri	1 Paheri	1 Rajuri
8	2 Cecil	1 Cecil	3 Cecil	2 Bomb.G.	2 Bomb.G.	3 Bomb.G.	2 Ameeri	3 Ameeri	2 Ameeri	1 Bennett	3 Bennett	1 Bennett	1 W. Lang.	1 W. Lang.	1 W. Lang.	2 Rajuri
9	3 E. Gold	3 E. Gold	2 E. Gold	1 EdXKent	1 EdXKent	3 EdXKent	4 Oakrong	2 Oakrong	3 Oakrong	3 H.Camb	3 H.Camb.	3 H.Camb.	2 Nam	2 Nam	4 Nam	1 Gadoeng
10	1 Mun	2 Mun	3 Duncan	2 Duncan	2 Duncan	2 Madoe	2 Madoe	3 Madoe	3 Florigon	3 Florigon	3 Florigon	3 Cambod.	3 Cambod.	2 Cambod.	2 Gadoeng	5 Gadoeng
11	1 Mun	2 Hindi	3 Hindi	3 Hindi	1 Simmon	1 Simmon	2 Simmon	1 Edward	1 Edward	2 Edward	2 Micong.	3 Micong.	2 Micong.	2 Aroaman	2 Aroaman	1 Aroaman
12	2 D.Haden	1 R.Saigon	1 R.Saigon	1 R.Saigon	1 J.Saigon	1 J.Saigon	2 J.Saigon	1 Glenn	4 Glenn	3 Glenn	2 EdXEd15	3 EdXEd15	2 EdXEd15	2 Golek	2 Golek	1 Golek
13	2 D.Haden	2 D.Haden	1 Keenan	1 Keenan	1 Keenan	1 Dixon	2 Dixon	5 Dixon	1 Martin	1 Martin	1 Martin	2 Brooks	2 Brooks	3 Brooks	4 Osteen	4 Osteen
14	3 Ryechk	3 Ryechk	1 Ryechk	1 San.6	1 San.6	2 San.6	3 G.Brooks	3 G.Brooks	2 G.Brooks	1 Webber	2 Webber	1 Webber	2 Parvin	2 Parvin	2 Parvin	4 Osteen
15	1 Smith	1 Smith	2 Smith	1 V. Pride	2 V. Pride	1 V. Pride	2 Keitt	2 Keitt	2 Keitt	2 Beverly	2 Beverly	2 Beverly	2 Sensa.	2 Sensa.	2 Sensa.	2 Ford
16	2 Jacq.	2 Jacq.	3 Andersn	2 Andersn	2 Andersn	1 Ruby	1 Ruby	1 Ruby	3 Spring.	2 Spring.	2 Spring.	3 Scupp.	3 Scupp.	4 Scupp.	2 Ford	2 Ford
17	4 Jacq.	3 Haden	1 Haden	1 Haden	2 Fascell	1 Fascell	1 Fascell	1 Kent	2 Kent	3 Kent	1 Pittigr.	1 Pittigr.	1 Pittigr.	1 Eldon	1 Eldon	2 Eldon
18	2 Harris S.	1 Lathrop	1 Lathrop	1 Lathrop	1 Earle1	1 Earle1	1 Earle1	1 Pruter	1 Pruter	1 Pruter	3 Irwin	2 Irwin	3 Irwin	4 Glazier	3 Glazier	3 Glazier
19	1 Harris S.	2 Harris S.	1 Cogshall	1 Cogshall	1 Cogshall	2 Zill	2 Zill	3 Zill	*	*	*	3 Reason.	1 Reason.	2 Reason.	2 Carrie	2 Carrie
20	1 Alice	1 Alice	1 Alice	2 Pills.A	1 Pills.A	2 Pills.A	4 Pills.	4 Pills.	4 Pills.	3 Gouvea	3 Gouvea	3 Gouvea	3 Momi K.	4 Momi K.	3 Momi K.	3 Carrie
21	2 Monser	2 Monser	2 Monser	1 Joellen	1 Joellen	1 Joellen	1 4 Way	2 4 Way	2 4 Way	2 Harris	2 Harris	2 Harris	3 Burgner	3 Burgner	3 Burgner	2 A.Reas.
22	2 Sunset	3 Sunset	5 Lucille	3 Lucille	2 Lucille	3 M. Late	2 M. Late	3 M. Late	3 Adams	2 Adams	2 Adams	3 Pope	3 Pope	3 Pope	1 A.Reas.	1 A.Reas.
23	2 Sunset	3 Lippens	3 Lippens	3 Lippens	1 Hansen	1 Hansen	1 Hansen	2 B.Herm.	2 B.Herm.	3 B.Herm.	3 Wallace	2 Wallace	3 Wallace	3 Hodson	2 Hodson	3 Hodson

Fig. 1. Disease ratings and arrangement of mango cultivars in germ plasm collection located at the University of Florida, Tropical Research and Education Center. All indicated trees were included in the study. Top of figure is orientated to north.

the disease in affected trees, symptom ratings reported here are those observed only in 1990, the most severe of the 3 years of observation.

Isolation and identification. Isolation procedures were slightly modified from those previously described (15). Tissues from trees displaying tip dieback symptoms were collected, surface-sterilized with 1% sodium hypochlorite for 3 min, and rinsed three times with sterile distilled water. Bark was removed, and sections from the vascular tissue were dissected with a sterile scalpel and placed on potato dextrose agar (PDA) or malt agar. Streptomycin sulfate was added to the first medium to prevent bacterial growth.

B. ribis anamorph *Fusicoccum* sp. was identified in the laboratory by its colony morphology, rapid growth rate, dark mycelium, and the formation of chlamydospores and immature conidial initials as previously described (15). Affected tissues produced stromatic pycnidia, which yielded hyaline, one-celled pycnidiospores. (15). When possible given the large number of samples, the isolates were grown side-by-side with our IMI 293744.

Statistical analysis. The plot design was dictated by the previously established arrangement of cultivars in the germ plasm collection. The tree replicates of each cultivar were planted adjacent to each other in

the collection (Fig. 1). The major systematic groups or embryonic types were randomly distributed throughout the collection. The General Linear Models procedure (GLM), nested analysis of variance (SAS Institute, Cary, NC), was used to compare disease responses among the major mango systematic groups and between the embryonic types. The type III *F* value, using MS for cultivar (nested with group), was used as an error term to compare disease responses among the major systematic groups. Similarly, the Type III *F* value, with MS representing cultivar (nested with embryo), was used as an error term to compare the two embryonic types. The *F* test with MSE as the error term was used to determine variability of disease responses among cultivars within major systematic groups and embryonic types. Embryonic type was regarded as a fixed effect; whereas cultivar was treated as a random effect (SAS).

The assumption of a homogeneous distribution of disease scores throughout the field was tested by fitting a second-degree polynomial regression model to the scores, based on row and column coordinates, with row and column indices as predictors. An *F* test was used to compare this model with one with only an intercept term. The disease scores were modeled as a function of row, row², column, column², and a

row-column interaction term. A finding of significance in any one of these terms indicated nonrandom distribution (SAS).

RESULTS

The mango cultivars used in this study were organized into eight major groups, as previously stated. Table 1 lists the cultivars assigned to each group and includes the embryony as well as the disease ratings for each cultivar. The lowest mean disease rating was found in the group formed by other *Mangifera* spp. (*M. odorata* and *M. zeylanica*), and the highest was found in the Turpentine types; however, when these results were compared with the other groups, no significant differences in severity of tip dieback disease were found (Tables 1 and 2) (*df* = 7, *F* value 1.10, *P* > 0.3690). To learn the effect of those cultivars free of the disease in the disease ratings of each major group, disease-free cultivars were excluded from the population. No significant differences were found among cultivars (*df* = 7, *F* = 0.88, *P* > 0.5262). There were, however, significant differences in disease severity among individual cultivars (*df* = 121, *F* value = 3.52, *P* > 0.0001).

A number of disease-free mango cultivars were found. Of the Indian monoembryonic group of cultivars, four out of 14 (29%) were disease free. Eight out of 46 in the Haden part (17%) and seven out of 23 in the Sandersha part (17%), both of the mainly monoembryonic group of Haden-Sandersha Complex, were free of the disease (Tables 1 and 2). The West Indian group, composed of monoembryonic and polyembryonic cultivars, displayed one out of seven (14%) disease-free cultivars. Only two out of 19 (10%) of the cultivars in the polyembryonic Southeast Asia group and none of the cultivars in the polyembryonic Turpentine group were disease free.

Because most of the disease-free cultivars were found to be monoembryonic, we tested the possibility that disease resistance was associated with embryony. Although 17% of the monoembryonic cultivars were free of the disease and only 8% were disease free in the polyembryonic population, no significant differences in mean tip dieback disease ratings were found between the two populations (Table 3). The mean tip dieback disease ratings of the monoembryonic and polyembryonic cultivars were 2.06 and 2.23, respectively (*df* = 1, *F* value = 1.40, *P* > 0.2388). We also considered the possibility that Haden-Sandersha Complex cultivars, which are composed of Florida and Hawaiian hybrids of Indian and Southeast Asian origin, may display intermediate characteristics. When these groups were removed from the data set, the means of dieback disease ratings were comparable to the previous analysis (1.99 for monoembryonic and 2.28 for polyembryonic cultivars). The analysis of variance of the *F* test reduced the *P* value; however,

Table 2. Severity of tip dieback disease of major groups, races or complexes of mango (*Mangifera indica* L.) and relatives in the germ plasm collection at the University of Florida, Tropical Research and Education Center, Homestead

Major group	No. of trees	No. of cultivars	Mean disease rating ^a		% Disease free cvs
			Total cvs	Diseased cvs	
Indian types	41	14	1.85 ± 0.15	2.21 0.17	29
Southeast Asia types	57	19	2.23 ± 0.13	2.30 0.13	5
West-Indian types	19	7	2.26 ± 0.25	2.50 0.25	14
Haden-Sandersha—Haden part	138	46	2.09 ± 0.08	2.32 0.08	17
Haden-Sandersha—Sandersha part	69	23	2.03 ± 0.11	2.25 0.12	17
Turpentine types	19	7	2.74 ± 0.24	2.74 0.24	0
Nonclassified cultivars	12	4	2.08 ± 0.29	2.08 0.29	0
<i>Mangifera</i> spp. (<i>M. odorata</i> and <i>M. zeylanica</i>)	6	2	1.67 ± 0.33	1.67 0.33	0

^a Mean disease ratings 1 to 5: 1 = free of tip dieback, 5 = severely damaged plants. GLM, nested. No significant differences were found among major races or groups of cultivars. Test hypothesis using the type III MS for cultivar (group) as an error term (*df* = 7, *F* value 1.10, *P* > 0.369). Groups or races were randomly distributed. The three replicated trees from each cultivar were not distributed at random.

Table 3. Severity of tip dieback disease as related to the embryony of cultivars in the germ plasm collection at the University of Florida, Tropical Research and Education Center, Homestead

Embryo type	Total population studied				Population of diseased cultivars ^b		
	No. of cultivars	No. of trees	Disease rating ^a	% disease free cvs	No. of cvs	No. of trees	Disease rating
Monoembryonic	86	256	2.06 ± 0.06	17	32	214	2.28 ± 0.06
Polyembryonic	36	105	2.23 ± 0.10	8	33	93	2.34 ± 0.10

^a Mean tip dieback disease ratings followed by standard errors.

^b Population excluding the disease-free cultivars. Ratings 1 to 5: 1 = free of tip dieback; 5 = severely damaged plants. No significant difference was found between the monoembryonic and polyembryonic populations of cultivar. GLM nested. Test the hypothesis using type III MS for cultivar (embryo) as an error term (*df* = 1; *F* value = 1.40, *P* > 0.2388). There was significant variability in disease response among cultivars within each embryo type. The *F* value using MSE as the error term was 3.54, *df* = 121, *P* > 0.0001.

differences in disease responses were not significant ($df = 1$, F value = 2.37, $P > 0.129$). There was significant variability in disease responses among the cultivars within each embryo type ($df = 121$, $F = 3.47$, $P > 0.0001$). To determine the effects of the cultivars that were free of the disease on the mean, the 17 cultivars free of disease (14 in the monoembryonic group and three in the polyembryonic group) were excluded from the computation (Table 3). The mean disease ratings were 2.28 and 2.34 for the monoembryonic and polyembryonic populations, respectively. No significant differences were found ($df = 104$, $F = 0.42$, $P > 0.5167$).

B. ribis anamorph *Fusicoccum* sp. was the only fungus found in high frequency in symptomatic tissues of diseased trees sampled in all areas of the orchard (data not shown). The incidence of recovery of *B. ribis* was 42% of the total number of symptomatic trees examined. This frequency of recovery was much higher than that of any other isolated fungus. *B. ribis* was not found in asymptomatic tissues. *Diplodia* sp. was also isolated from diseased trees but in much lower frequency, as were other previously reported secondary fungi (15).

The disease scores appeared to be uniformly distributed throughout the field (Fig. 1). High and low disease scores were found in most areas. The assumption of a homogenous distribution of the scores was not supported after examination of the second-degree polynomial regression model to the scores based on row and column coordinates. This second-degree model fit significantly better ($F = 5.10$; model $df = 5$, error $df = 349$; $P > 0.0002$) than did the model with only an intercept term ($P > 0.0002$), indicating that similar disease scores were not uniformly distributed across the field. The F test indicated significance between rows ($P > 0.025$) and no significance between columns ($P > 0.146$) or in the interaction between rows and columns ($P > 0.573$).

DISCUSSION

The primary objective of this study was to investigate the possibility that disease resistance to mango tip dieback is associated with one or more major mango races, groups, or complexes described in earlier systematic studies (14,16). No attempt was made to group the cultivars according to other comparable phylogenetic studies (2,5,20) because they did not include the cultivars or *Mangifera* species observed in the present research. A consistently high incidence of disease was found throughout the population of trees. *B. ribis* was isolated from trees in all sections. A small number of individual trees among the three replicate trees of some cultivars escaped infection. Although no significant differences in the mean disease ratings of tip dieback disease were found among the

systematic groups of mango cultivars or relatives, there were distinct differences in the proportion of cultivars found to be disease free in the various groups. Most of the cultivars found to be free of disease occurred in groups dominated by monoembryonic types (Table 1). The exclusively monoembryonic Indian group displayed the highest proportion of disease-free cultivars; whereas the groups composed exclusively of polyembryonic types, such as the Southeast Asia and Turpentine groups, displayed the lowest proportion (Table 2).

Although the proportion of disease-free cultivars in the monoembryonic types was twice as high as that of the polyembryonic types, no difference in the mean disease rating was found between the two types. High disease ratings found in susceptible monoembryonic cultivars offsetting the higher proportion of disease-free cultivars found in that type may account for the similar means.

It is plausible that a greater proportion of the monoembryonic population of cultivars was resistant to tip dieback disease than were the polyembryonic types, because of the greater probability of resistance introduction in the former through sexual reproduction (3,8). Zygotic embryos of monoembryonic cultivars would theoretically have the greater evolutionary advantage in developing resistance to the pathogen than would the adventive, asexual embryos in seeds of polyembryonic types. The latter are derived from the nucellar inner integument of the ovule (10,17). Although Sturrock (23) considered polyembryony to be the more primitive character, and some reversion in embryony has been observed (23), Kostermans and Bompard (9) listed only three out of 69 *Mangifera* species (*M. indica*, *M. casturi*, and *M. laurina*) where polyembryony occurs. It is thus difficult to accept Sturrock's original contention. Rhodes et al. (16) found good separation of phenotypic characters between monoembryonic and polyembryonic cultivars. There was substantial variability in disease responses among the cultivars within each embryony type.

One source of variability of disease rating in the major mango races may be derived from the large number of mango hybrids of intermediate nature within the Florida and Hawaii Haden-Sandersha Complex. Many of these mango cultivars have been developed from seedlings of natural crosses between the diverse monoembryonic and polyembryonic types whose centers of origin are India and Southeast Asia, respectively. As observed by Rhodes et al. (16) in their systematic phylogenetic work, the inclusion of these hybrids exerted an intermediate effect on the clustering of major groups. Removal of the Haden-Sandersha Complex cultivars from the data set decreased the variability displayed by the two embryonic types but

was not sufficient to result in a statistical difference between them. There was also a lack of significant difference between the monoembryonic Indian and polyembryonic Southeast Asia groups (Table 2).

B. ribis was consistently isolated from symptomatic tissues of diseased trees located in most areas of the orchard (data not shown). The disease scores were apparently uniformly distributed throughout (Fig. 1), and high and low disease scores were found in most areas, suggesting that the fungus inoculum was available in all sections of the orchard. Nevertheless, the assumption of a homogeneous distribution of disease scores was not supported by the statistical test in rows, and the hypothesis of uniformity was rejected. Significant differences were found among rows, but no significant differences were found among columns or in the interaction of rows and columns. These findings suggest that differences attributed to cultivars may actually be influenced by conditions in the row, but most likely suggest that indices of the rows may be influenced by the clustering of the three replications of each mango cultivar in the rows. The disadvantages of the preset conditions with the three non-random replications of each cultivar within rows were mentioned previously.

The study presented here provides the first indication that field resistance to tip dieback disease may be present in some mango cultivars. These results are valuable for development of cultivars and future research; efforts should include the evaluation of those cultivars showing field resistance using controlled conditions.

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