Neofusicoccum mangiferae (Crous et al., 2006)

Synonyms
Dothiorella mangiferae, Fusicoccum mangiferae, Hendersonula cypria, Nattrassia mangiferae, Neofusicoccum mangiferum

Common Name(s)
Mango fruit rot, Stem-end rot, inflorescence blight of mango

Type of Pest
Fungal pathogen

Taxonomic Position
Class: Dothideomycetes, **Order:** Botryosphaeriales, **Family:** Botryosphaeriaceae

Reason for Inclusion in Manual
Suggestion from CAPS community; recent pest find in Puerto Rico

Figure 1. Symptoms of fruit rot in mango caused by *N. mangiferae*. Photo courtesy of Hui-Fang Ni.

Background Information
Neofusicoccum mangiferae and related fungi are collectively the causal agents of mango fruit rot/ stem-end rot. These fungi are either pathogenic or endophytic inhabitants of plant hosts. *N. mangiferae* infections can be symptomatic or asymptomatic, which makes detection difficult and inconsistent (Heath et al., 2011). *N. mangiferae* causes dieback and fruit rot/ blossom decline on several fruit and ornamental tree species. Most notably, it causes decline and dieback of mango trees (Heath et al., 2011). In addition, *N*. *mangiferae* causes pre- and post-harvest infection of mango fruit (Fig. 1), inflorescence blight and rachis necrosis of mango (Fig. 3) (Serrato-Diaz et al., 2014) and also causes avocado fruit rot (Fig. 4) (Ni et al., 2009). Stem-end rot of fruit typically appears post-harvest and can severely affect fruit quality (Ploetz et al., 1994).

Pest Description
Neofusicoccum mangiferae produces colonies of gray, felted mycelium with partly immersed, discrete conidiomata on oatmeal agar and radially dendritic, dark gray mycelium with a "pepper-spot" pattern of pycnidial 3 initials on Potato Dextrose Agar (PDA) within 7-10 days (Fig. 2). On mango, unilocular conidiomata are immersed in a subcuticular pseudostroma. Conidia (9-18 × 4-6 µm) are ellipsoid to ovoid (Fig. 2), hyaline, aseptate, and shorter and broader than those of *Dothiorella dominicana*.

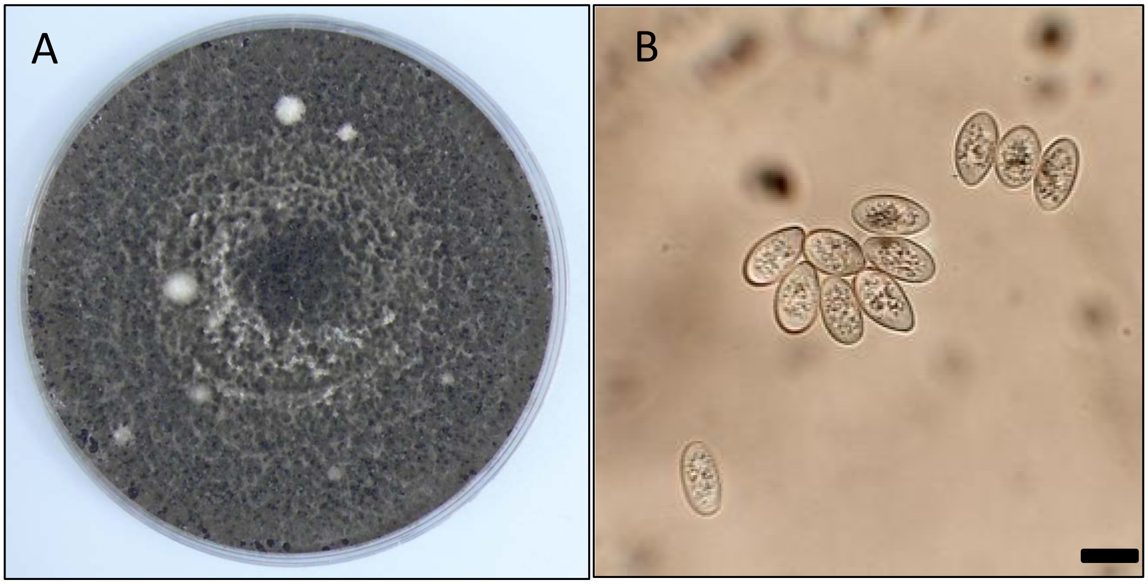

Figure 2. A. Morphology of *N. mangiferae* cultured on PDA for 10 days. Courtesy of Hui-Fang Ni. **B.** Conidia of *N. mangiferae*. Bar= 10µM. Photos courtesy of Yei-Zeng Wang.

Conidia obtained from pycnidia were ovate, one-celled, and hyaline, with an average length and width of 12.93 ± 0.93 × 6.98 ± 0.40 µm and an average length/ width ratio of 1.85 (Ploetz et al., 1994; Ni et al., 2010) (Fig. 2). Two-septate, brown-walled conidia and microconidia (2.5 × 4.5 µm) are sometimes observed (Ploetz et al., 1994). A distinguishing feature between *N*. *fusicoccum* and *Neoscytalidium dimidiatum* is the type of synanamorph each produces (Crous et al., 2006; Romberg, 2013). *Neofusicoccum mangiferae* produces a *Dichomera*-like synanamorph with brown, globose to pyriform conidia (Crous et al., 2006). This pathogen can be isolated on agar and on host tissue and produces distinct spores and growth patterns (Ploetz et al., 1994).

Fungal colonies from avocado that were cultured on acidified PDA (with lactic acid; pH 3.8) were initially colorless, turned dark gradually, and ultimately became gray to dark gray (Fig. 2). After 4 days under fluorescent light at 25°C (77^{o}F), pycnidia formed on PDA. Conidia obtained from fruiting bodies were ovate, one-celled, and hyaline, with an average length and width of 12.9 (9.9 to 15.6) × 6.4 (5.2 to 7.2) µm (Ni et al., 2009).

Colonies on Malt Extract Agar (MEA) rapidly spread, covering an entire petri dish in 7 days at room temperature. Mycelium is grayish-white at first, turning olivaceous-brown, floccose, forming scattered, grayish rope-like strands. Pycnidia superficial, globose to pyriform, ostiolate, 120-200 µm wide, sometimes aggregated up to 1 mm wide, blackish brown. Conidia hyaline, broadly ellipsoid with a flat base, smooth, 12-15 x 7-8 µm (L/B = 1.75) (Huang and Wang, 2011).

Biology and Ecology

The pathogen occurs in mature stem tissue and infects fruit from endophytically colonized inflorescence, pedicle, and peduncle tissues. Water stress predisposes host

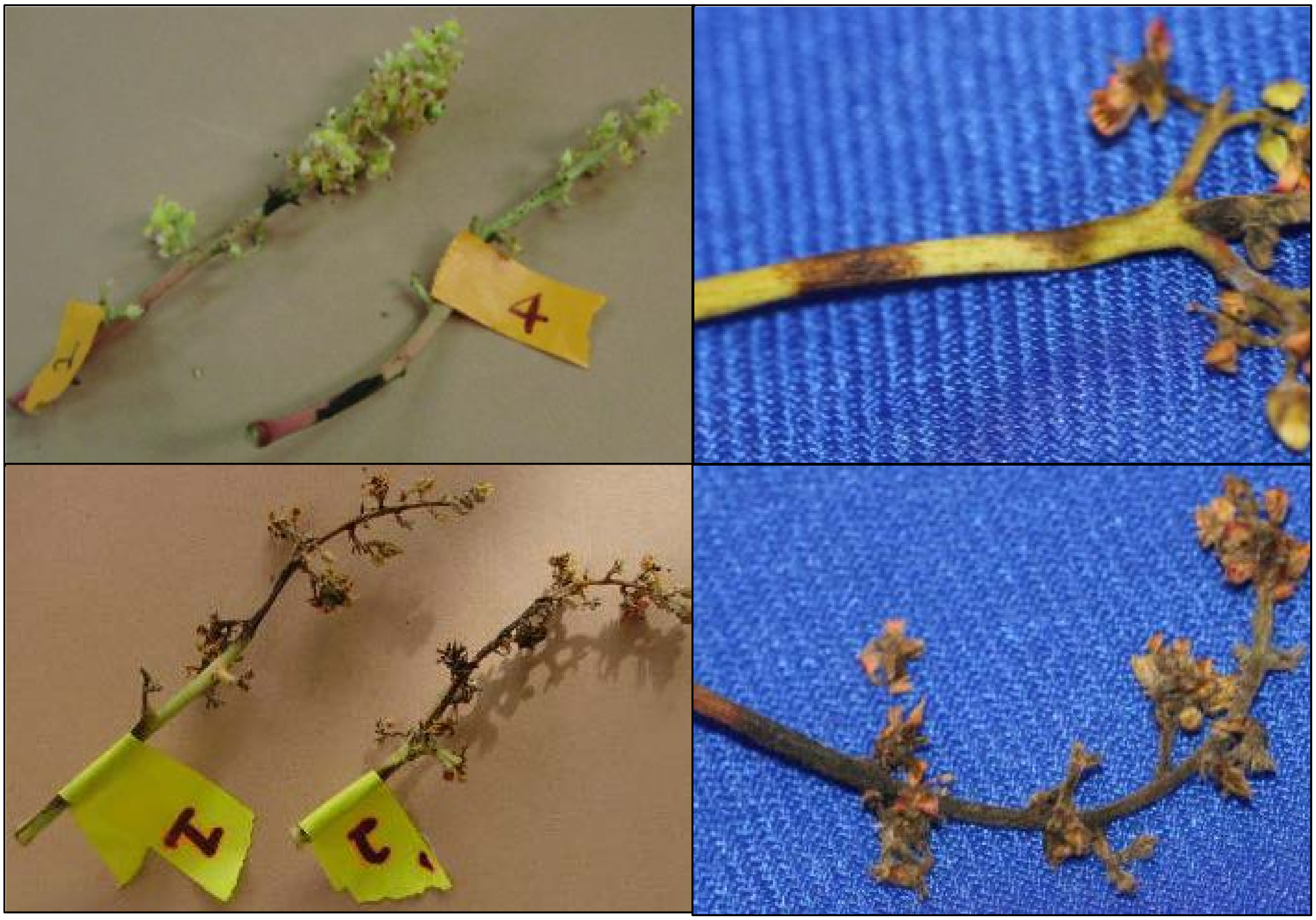

Figure 3. Symptoms of *N. mangiferae* infection on mango inflorescences**. Top.** Rachis necrosis*.* **Bottom.** Inflorescence blight caused by *N. mangiferae*. Courtesy of Luz M. Serrato-Diaz.

plants to endophytic infections. These infections are not apparent until fruit development is well advanced, making detection possible in mango stems, pedicels, and fruit during fruit maturation. Detection may also be influenced by the presence of other fungi (Johnson et al., 1994). Thus, stem-end rot is called a post-harvest disease (Ploetz et al., 1994). However, pre-harvest infections can also occur through wounds, but like the endophytic infections, these infections are latent until the fruit is harvested and matured (Ploetz et al., 1994).

Spores of *Neofusicoccum* spp. are produced in tree litter and on dead leaves, twigs, and branches in the tree canopy. These spores are spread by water and air movement (Ploetz et al., 1994). Stem-end rots of mango fruits are affected by periods of rain and high relative humidity at the beginning and ending of a dry season (Prusky et al., 2009) and are often triggered by prolonged storage at cool temperatures and water stress in the host plants (Ploetz et al., 1994). When isolated from mango trees with branch wilt, the optimal temperature for *N*. *mangiferae* in culture was 35°C (95^{o}F) with no growth occurring at 40°C (104^{o}F) (El-Trafi, 2009).

Losses from stem-end rot fungi of mango increase when fruit are stored for prolonged periods at low temperature or when fruit ripen at temperatures of 28°C (82°F) or greater (Ploetz et al., 1994).

Symptoms/Signs

In Mango: *Neofusicoccum mangiferae* produces "tearstained" patterns and localized lesions that appear as discrete, dendritic, superficial spots on fruit (Fig. 1) (Ploetz et al., 1994). Necrosis remains beneath the cuticle and penetrates the entire fruit within several days at 25°C (77°F) (Johnson et al., 1991). Superficial mycelium appears around the pedicel or through ruptures in the skin or directly through the epidermis. A brown liquid (exudate) can be observed oozing from the stem end or from surface ruptures (Johnson et al., 1991). Stem-end rot fungi in mango are also associated with twig and branch dieback. In mango hosts, inflorescence blight showing rachis and flower necrosis is also present (Fig. 3) (Serrato Diaz et al., 2014).

In Avocado: Infected avocados developed smooth, brown, circular spots first on the surface of harvested fruits (Fig. 4). *Neofusicoccum mangiferae* was isolated from the margin of avocado lesions and could also be found from symptomless fruit pedicles and stems (Ni et al., 2009).

Pest Importance

Both avocados and mangos are produced in tropical and subtropical portions of the United States. In the United States, avocado production acreage was 64,700 acres in 2013 (California with 57,300 acres, Florida with 7,000 acres, and Hawaii with 400 acres). Puerto Rico had 32,591 avocado trees of bearing acreage and 16,279 trees of non-bearing acreage (USDA, 2012a). Mango-bearing and non-bearing acreage in the United States as of 2012 was 3,006 acres, all in California, Florida, Hawaii, and Texas. The majority of this acreage is located in Florida (USDA, 2012b; USDA-NASS, 2015).

Figure 4. Symptoms of fruit rot in avocado caused by *N. mangiferae*. Courtesy of Hui-Fang Ni.

In 2012, mango was grown in Puerto Rico on 157 farms, and 167,130 trees of bearing age were present on the island (USDA, 2012a).

Neofusicoccum mangiferae is not specifically reported as a harmful organism by other countries (PExD, 2015). However, its synonym *Nattrassia mangiferae* is reported as harmful by Ecuador. *Dothiorella* spp. are reported as harmful by Israel, and *Fusicoccum* spp. (former synonomous genera) are considered harmful by South Korea (PExD, 2015).

Known Hosts

Due to the considerable changes that have taken place recently in the taxonomy of the Botryosphaeriaceae, older literature addressing patterns of diversity and host association must be interpreted with caution (Slippers and Wingfield, 2007). The host range of *N. mangiferae* is presented here based on current synonyms.

Major hosts: *Mangifera indica* (mango) and *Persea americana* (avocado).

Other hosts: *Agathis robusta* (Queensland kauri pine), *Dioscorea rotundata* (white Guinea yam), *Ficus religiosa* (sacred fig), *Manihot esculenta* (Brazilian arrowroot, cassava), *Plumeria obtusa* (Plumeria), *Prunus armeniaca* (apricot), *Psidium guajava* (guava), *Syzgium cordatum* (umdoni, waterberry), and *Tibouchina* spp. (tibouchina) (Crous et al., 2006; Ni et al., 2009; Heath et al., 2011; NPAG, 2015)

Known Vectors (or associated insects)

Neofusicoccum mangiferae is not known to be vectored by any organism.

Known Distribution

Africa: Benin, Nigeria, South Africa. **Asia:** India, Iran, Myanmar, Pakistan, Taiwan, Thailand. **Europe:** Cyprus. **North America:** Puerto Rico. **Oceania:** Australia (El-Trafi, 2009; Ni et al., 2009; Heath et al., 2011; Farr and Rossman, 2013; Serrato-Diaz et al., 2014; NPAG, 2015).

There have been reports of *N. mangiferae* in the continental United States, but none have been verified by examination of a maintained culture or DNA sequences. These reports have been determined to be misidentifications or non-verifiable (Romberg, 2015, personal communication). In 2014, there was an interception of *N. mangiferae* in California on mango fruit in a mail shipment from Florida. The pathogen, however, was not confirmed on the tree where the fruit reportedly originated (Romberg, 2015, personal communication).

Pathway

Currently, open pathways include propagative material of *Agathis* spp. and *Plumeria* spp. imported from countries where the pathogen has been recorded (USDA, 2015). *Neofusicoccum mangiferae* has been isolated from asymptomatic tissue of the ornamental shrubs and trees of *Tibouchina*, which suggests it can exist latently or as an endophyte (Heath et al., 2011) and, therefore, may move undetected in plants (NPAG, 2015).

Mango fruit for consumption is permitted entry from multiple countries with records of the pathogen. The phytosanitary measure in place requires a certificate issued by the national plant protection organization of the country with either additional or no additional declarations (FAVIR, 2014).

There has been one interception of mangoes (*Mangifera* spp.) in Hawaii originating from India (2012) and one from China (2011). There has also been one *M. indica* interception in Hawaii originating from India (1994) (AQAS, 2015).

Since 2013, there have been interceptions of *Neofusicoccum* spp. from Thailand (1) and Japan (1) on host material intended for propagation (AQAS, 2015). The Thailand interception was on *Plumeria* spp., a known host of *N. mangiferae*, so this interception could have been *N. mangiferae*.

Potential Distribution within the United States

From NPAG, 2015: The pathogen has been reported in areas corresponding to Plant Hardiness Zones 9 to 13, with possibly some occurrence in Zone 8. This matches the climate preferences for the hosts listed in the host range section, including *Tibouchina urvilleana* (DC.) Cogn., which will grow in Zones 8-12 (Scheper, 2005). Although freezing temperatures in Zone 8 will kill the plants to the soil surface, "they usually resprout with the return of warm weather and recover to bloom in mid to late summer" (Scheper, 2005). *Neofusicoccum mangiferae* in culture prefers to grow at 35°C (95^{o}F) and will not grow at 40°C (104^{o}F) (El-Trafi, 2009), but we found no information on lower temperature limits for this pathogen, and it is unknown if the pathogen will be limited by the distribution of known hosts. Thus, N. *mangiferae* may become established in Plant Hardiness Zones 9 to 13 of the United States and possibly Zone 8, which correspond to the southern United States, island states and territories, and possibly the Pacific Northwest.

California, Hawaii, Florida, and Puerto Rico, are all vulnerable to *N. mangiferae* establishment due to the commercial production of avocado and/or mango in those states/territories (USDA, 2012ab; USDA-NASS, 2015). All other mango producing islands or territories (for local or homeowner consumption) may be vulnerable from importation of mangos or other susceptible fruits from known distribution areas for the pathogen.

Survey

Approved Method for Pest Surveillance*:

The approved survey method is to collect symptomatic plant tissue by visual survey.

*For the most up-to-date methods for survey and identification, see Approved Methods on the CAPS Resource and Collaboration Site, at https://caps.ceris.purdue.edu/approved-methods.

Literature-Based Methods:

Visual symptoms in mango and avocado hosts are visible in infected inflorescenses and fruit, (Ni et al., 2009; Ni et al., 2010; Heath et al., 2011; Serrato Diaz et al., 2014), but they may be confused for symptoms of other Botryosphaeriaceae species pathogens.

There are several fungal pathogens of mango which can be easily confused for *N. mangiferae* (Ni et al., 2012), and mixed infections of *N. mangiferae* and other fungal pathogens are common (Serrato-Diaz et al., 2014).

Key Diagnostics

Approved Method for Pest Surveillance:

The approved method for *N. mangiferae* identification is morphological. Ni et al. (2009) describe the morphological features of this fungus when cultured on PDA (Ni et al., 2012).

Note: *Neofusicoccum mangiferae*, if care is taken, is relatively easy to identify using morphological characters (McKemy, 2015, personal communication),

Although molecular methods are not validated, there are molecular methods available in the literature for confirmation.

A morphological key is also available at: Key to the various lineages in "Botryosphaeria" to assist with a morphological identification.

*For the most up-to-date methods for survey and identification, see Approved Methods on the CAPS Resource and Collaboration Site, at https://caps.ceris.purdue.edu/approved-methods.

Literature-Based Methods:

Ni et al. (2010) identified *N. mangiferae* by performing PCR amplification by universal primers, ITS1/ITS4, and sequenced the ribosomal DNA of the internal transcribed spacer (ITS1-5.8S-ITS2 rRNA gene cluster).

Ni et al. (2012) developed a nested PCR which can distinguish *N. mangiferae* from other Botryosphaeriaceae species.

Serrato-Diaz et al. (2014) describe a molecular method for identification by PCR amplification and sequencing of the Internal Transcribed Spacer (ITS) rDNA.

Easily Confused Species

There are several fungal pathogens of mango which can be easily confused for *N. mangiferae*. For example, *Lasiodiplodia theobromae*, *Neofusicoccum parvum*, and *Fusicoccum aesculi* are all anamorphs of Botryosphaeriaceae species and are also the causal agents of mango stem-end rot and fruit rot. *Lasiodiplodia theobromae* and *N. parvum* are both present in Puerto Rico (Serrato-Diaz et al., 2013ab). In addition, numerous other fungal pathogens are known to infect mango, including: *Phomopsis* spp., *Colletotrichum gloeosporioides*, *Fusarium* spp., *Pestalotiopsis* spp., *Alternaria* spp., *Guignardia* spp., *Cladosporium* spp., *Ceratocystis* spp., and *Botrytis* spp. (Ni et al., 2012).

Several other species of Botryosphaeriaceae have also been reported on avocado, including *N. parvum*, *F. aesculi*, and *Dothiorella aromatic* (Ni et al., 2009).

References

AQAS. 2015. Agricultural Quarantine Activity Systems. Queried March 9, 2015 from, https://aqas.aphis.usda.gov/aqas/

BONAP. **2014.** Biota of North America Program. Queried 4/29/15 from, www.bonap.org/

Crous, P. W., B. Slippers, M. J. Wingfield, J. Rheeder, W. F. O. Marasas, A. J. L. Philips, A. Alves, T. Burgess, P. Barber, and J. Z. Groenewald. 2006. Phylogenetic lineages in the Botryosphaeriaceae. Studies in Mycology 55: 235-254.

El-Trafi, M. A. 2009. Studies on mango branch wilt disease caused by *Neofusicoccum mangiferae* (Abst.). Sudan Academy of Sciences, Khartoum, Sudan.

Farr, D. F., and A. Y. Rossman. 2013. Fungal Databases - *Neofusicoccum mangiferae*. Systematic Botany and Mycology Laboratory, ARS, USDA. Last accessed December 10, 2013, from http://nt.ars-grin.gov/fungaldatabases/

FAVIR. **2014.** Fruits and Vegetables Import Requirements**.** USDA-APHIS, retrieved from, http://www.aphis.usda.gov/favir/

Heath, R. N., J. Roux, B. Slippers, A. Drenth, S. R. Pennycook, B. D. Wingfield, and M. J. Wingfield. 2011. Occurrence and pathogenicity of *Neofusicoccum parvum* and *N. mangiferae* on ornamental *Tibouchina* species. For. Path. 41: 48–51.

Huang, C. L., and Y. Z. Wang. 2011. New Records of Endophytic Fungi Associated with the Araucariaceae in Taiwan. Collection and Research 24: 87-95.

Johnson, G. I., A. W. Cooke, A. J. Mead, and I. A Wells. 1991. Stem end rot of mango in Australia: causes and control. Acta Horticulturae 291:3.

Johnson, G. I., A. J. Mead, A. W. Cooke, and I. A. Wells. 1994. Stem-end rot diseases of tropical fruit - mode of infection in mango, and prospects for control. Pages 72-76 ACIAR Proceedings: No. 58.

McKemy, J. 2015. RE: *N. mangiferae* post assessment. Personal communication to Melinda Sullivan on March 17, 2015, from Dr. John McKemy, USDA APHIS PPQ National Mycologist/ Plant Pathologist.

Ni, H. F., R. F. Liou, T. H. Hung, R. S. Chen, and H. R. Yang. 2009. First report of a fruit rot disease of avocado caused by *Neofusicoccum mangiferae*. Plant Disease 93(7): 760.

Ni, H. F., R. F. Liou, T. H. Hung, R. S. Chen, and H. R. Yang. 2010. First report of fruit rot disease of mango caused by *Botryosphaeria dothidea* and *Neofusicoccum mangiferae* in Taiwan. Plant Disease 94(1): 128.

Ni, H. F., H. R. Yang, R. S. Chen, T. H. Hung, and R. F. Liou. 2012. A nested multiplex PCR for species-specific identification and detection of Botryosphaeriaceae species on mango. European Journal of Plant Pathology 133(4), 819-828

NPAG. **2015.** NPAG Report: *Neofusicoccum mangiferae* (Sydow & P. Sydow) Crous, Slippers & A.J.L. Phillips: Stem-end rot. USDA-APHIS PPQ-CPHST-PERAL, New Pest Advisory Group (NPAG), Raleigh, NC.

PExD. 2015. Phytosanitary Export Database. United States Department of Agriculture, queried April 1, 2015.

Ploetz, R. C., G. A. Zentmeyer, W. T. Nishijami, K. G. Rohrbach, and H. D. Ohr. 1994. Compendium of Tropical Fruit Diseases, St Paul, MN.

Prusky, D., I. Kobiler, I. Miyara, and N. Alkan. 2009. Fruit Diseases, 7. Pages 210-230. The Mango: botany, production and uses (2nd). CABI. CAB International, Cambridge, MA.

Romberg, M. 2015. RE_ *Neofusicoccum mangiferae* – status. Personal communication to Daniel Mackesy on April 2, 2015, from Dr. Megan Romberg, USDA APHIS PPQ National Mycologist/ Plant Pathologist.

Scheper, J. 2005. #109 *Tibouchina urvilleana*. Florida data. Retrieved from, http://www.floridata.com/ref/t/tibo_urv.cfm.

Serrato-Diaz, L. M., M. Perez-Cuevas, L. I. Rivera-Vargas, and R. D. French-Monar. 2013a. First report of *Lasiodiplodia theobromae* causing inflorescence blight of mango. Plant Disease 97(10): 1380.

Serrato-Diaz, L. M., M. Perez-Cuevas, L. I. Rivera-Vargas, and R. D. French-Monar. 2013a. First report of *Neofusicoccum parvum* causing rachis necrosis of mango (*Mangifera indica*) in Puerto Rico. Plant Disease 97(10): 1381.

Serrato-Diaz, L. M., L. I. Rivera-Vargas, and R. D. French-Monar. 2014. First report of *Neofusicoccum mangiferae* causing rachis necrosis and inflorescence blight of mango (*Mangifera indica*) in Puerto Rico. Plant Disease 98(4): 570.

USDA. 2012a. Puerto Rico Island and Municipo Data. *in*. National Agricultural Statistcal Service, Census of Agriculture.

USDA. 2012b. U.S. Mango-acres bearing and non-bearing and number of operations with bearing and non-bearing acreage by State and National total. *In*. National Agricultural Statistical Service, Census of Agriculture.

USDA. 2015. Plants for Planting Manual. Animal and Plant Health Inspection Service, Plant Protection and Quarantine (PPQ) Manuals Unit, Frederick, MD. Last updated January 21, 2015. http://www.aphis.usda.gov/import_export/plants/manuals/ports/downloads/plants_for_planting.pdf

USDA-NASS. 2015. Noncitrus fruits and nuts 2014 summary. United States Department of Agriculture, National Agricultural Statistics Service (NASS). http://usda.mannlib.cornell.edu/usda/current/NoncFruiNu/NoncFruiNu-01-23-2015.pdf

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