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ABSTRACT

Despite considerable research globally on postharvest diseases of mango, virtually no work has been done to determine the occurrence of these diseases and their causal agents in Fiji. This study is the first major field and market survey of postharvest diseases of mango in Fiji. For the field survey, fruits from five local mango cultivars were harvested at each of five locations. Fruits from five imported cultivars were also harvested from one location in Nadi. For the market survey, vendors were selected at five municipal markets for fruit collection, along with ten roadside stalls. For field and market surveys, fruits were incubated at 23°C and assessed for postharvest disease (incidence and severity of body rots and stem end rots) when ripe. Isolations were made from disease lesions and fungal cultures were identified using multilocus sequence typing. High incidences of body and stem end rots were recorded across all surveys. In the field survey of local cultivars, 'Salusalu' fruits were found to have the lowest severity of body rot and stem end rot on average, while for imported cultivars, 'Nam Doc Mai' had the lowest average severity of these diseases. The market survey showed that 'Salusalu' had a lower incidence of anthracnose on the body of fruit compared to all other cultivars, although it did have a surprisingly high incidence of stem end rot at some localities. Fungal isolates from anthracnose lesions in the field and market surveys were identified as *Colletotrichum asianum*, *C. simmondsii* and *C. fructicola*. *C. asianum* was the predominant species associated with anthracnose symptoms, accounting for 97% of isolations. *Lasiodiplodia theobromae* and *Neofusicoccum parvum* were the predominant species found in association with mango stem end rot symptoms. Other species isolated from mango stem end rot in lesser numbers included *L. brasiliensis*, *N. umdonicola* and *N. kwambonambiense*. All of these fungi represent new reports for Fiji.

Introduction

Fiji is a tropical island country with a climate highly suitable for mango production. While Fiji's domestic mango production is relatively small (260 metric tonnes in 2021;

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FAOSTAT 2023), mango is an important horticultural crop (Baker 2015). The Fiji mango season normally last for four months from October to January (Baker 2015) and is based on around 70 local and imported cultivars (Brown 2010). Most of Fiji's mangoes are grown in the drier north-west region of Viti Levu Island from Nadi to Rakiraki, and the northern parts of Vanua Levu Island. Mango is also grown in the south-east of Viti Levu Island around Suva, which has a relatively high rainfall (mean annual rainfall in excess of 3000 mm), and hot and humid conditions. In these wetter locations production is limited to a few cultivars such as 'Salusalu' (also known as 'Baramasia'). Most of the Fiji's mango production are sourced from small plantings grown near homesteads, along village boundaries and along roadsides. There is limited orchard management practice with planting based on locally sourced cultivars. Currently there are only a small number of commercial mango orchards in Viti Levu Island, primarily growing imported cultivars.

Internationally, mango production tends to be concentrated in the dry tropics (Johnson 2008), where postharvest diseases such as anthracnose are less problematic. When grown in humid, wet regions, anthracnose (caused by *Colletotrichum* spp.) limits productivity, reduces fruit quality and directly affects fruit marketability of mango fruits (Lima et al. 2013; Prusky et al. 2022). In fact, postharvest losses due to anthracnose can reach almost 100% of the harvested crop when fruits are grown under wet or very humid conditions (Arauz 2000).

Stem end rot, caused by a range of fungi (including several species in the family Botryosphaeriaceae), is another common postharvest disease of mango worldwide. Stem end rot tends to be more prevalent in fruits harvested from older trees, in stored fruits, and in situations where anthracnose has been well controlled (Johnson 1994).

Taxonomic revisions of the genus *Colletotrichum* (Phoulivong et al. 2010; Weir et al. 2012) and the family Botryosphaeriaceae (Slippers et al. 2017) have resulted in recognition of many new species. For the genus *Colletotrichum*, previously recognised species such as *C. gloeosporioides* and *C. acutatum* are now considered species complexes with many newly recognised species within. *Colletotrichum asianum*, the most common species found in association with mango anthracnose worldwide, is one such species within the *C. gloeosporioides* species complex. A number of other species within the *C. gloeosporioides*, *C. acutatum* and *C. boninense* species complexes have also been reported in association with mango anthracnose in various countries, (Lima et al. 2013; Honger et al. 2014; Liu et al. 2017; Qin et al. 2017). For the Botryosphaeriaceae, a number of species in the genera *Lasiodiplodia*, *Neofusicoccum, Botryosphaeria, Neoscytalididium* and *Pseudofusicoccum* have been reported as pathogens of mango, causing fruit rots, diebacks and stem cankers (Slippers et al. 2005; Pavlic et al. 2008; Abdollahzadeh et al. 2010; Sakalidis et al. 2011; Ismail et al. 2012; Ni et al. 2012; Marques et al. 2013; Phillips et al. 2013; Trakunyingcharoen et al. 2014; Li et al. 2021). *Lasiodiplodia theobromae* and *Neofusicoccum parvum* are however the most common species reported as causal agents of stem end rot in mango in many countries. Other fungi, not in the Botryosphaeriaceae, have also been reported as causal agents of stem end rot in mango, such as *Phomopsis mangiferae, Cytosphaera mangiferae* and *Pestalotiopsis mangiferae* (Johnson 2008).

While there has been extensive international research on postharvest diseases of mango, almost no work has been done in Fiji to determine the principal postharvest diseases of mango and their causal agents. There has also been no research undertaken to estimate the occurrence of these diseases on different cultivars and at different localities

in Fiji. To better understand the key postharvest diseases affecting mango in Fiji, the aim of this study was to assess the occurrence of postharvest diseases in mango, and identify their principal causal agents, in a range of local and imported cultivars growing in the principal growing regions of Viti Levu. This was done by harvesting fruits directly from mango plantings and assessing fruits for disease during the ripening process (field survey), and also by sourcing fruits from municipal fruit markets and roadside stalls at key localities for postharvest assessment (market survey). For the field and market surveys, fungal isolates obtained from symptoms on fruits were identified based on multilocus sequence typing.

Materials and methods

Field survey of local mango cultivars

A survey of mango postharvest diseases was conducted during the 2015/16 season at the five main mango production areas of Viti Levu (Nadi, Lautoka, Ba, Tavua, Rakiraki). At each locality, 30 fruits from each of five local cultivars ('Maqo Dina', 'Maqo Uto', 'Maqo Loa', 'Peach', 'Salusalu') were sourced from unsprayed trees. A total of 750 fruits were harvested across the survey. The trees sampled were mainly growing near homesteads and not cultivated commercially. Most had not received any agronomical inputs such as fertiliser, pesticide or herbicide application, or practices such as pruning.

Harvested fruits were transported to the laboratory in Koronivia, where they were de-sapped by trimming the pedicel to 5mm to allow sap to drain out for 30 min. Fruits were then washed with warm tap water and mild detergent to remove sap residues. Fruits were air-dried, placed in ripening boxes and stored in the laboratory at 23°C. Fruit firmness was monitored daily using gentle hand pressure on a 1–5 scale (where 1 = hard green fruit; 2 = first detectable softening; 3 = firm ripe; 4 = eating ripe; 5 = over-ripe) as described in Hassan (2006). When judged to be at the eating-ripe stage (rating 4), fruits were removed from storage and assessed for postharvest disease development and skin colour. All diseases occurring at the stem end of fruit (i.e. those caused by either *Colletotrichum* spp. or Botryosphaeriaceous fungi) were assessed collectively as 'stem end rot', and all diseases occurring on the body of fruits were similarly assessed as 'body rots'. The severity of body and stem end rots were estimated visually as the percentage of fruit surface area affected (external symptoms). Fruit skin colour was assessed visually on a 1–6 scale where: 1 = 100% green; 2 = colour break; 3 = 25% yellow; 4 = 50% yellow; 5–75% yellow; 6 = 100% yellow (Hassan 2006). Fruit shelf-life was defined as the number of days for fruits to reach the eating ripe stage at 23°C.

Field survey of imported cultivars

A second field survey was carried out at Legalega Research Station (Nadi) during the 2016/17 mango season. This site was the only locality where multiple imported cultivars were available. Thirty fruits from each of five imported mango cultivars ('Tommy Atkins', 'Kensington Pride', 'Tahitian', 'Nam Doc Mai' and 'Nang Klan Wan') and one local cultivar ('Maqo Dina') were harvested and assessed for postharvest disease development, skin colour and shelf-life as previously described. A total of 180 fruit were assessed.

Market survey of mango postharvest diseases in Fiji

The market survey focussed on the urban municipal markets of Ba, Lautoka, Rakiraki, Suva and Nausori, as well as roadside stalls along the Queens Road highway between Sigatoka and Lautoka. Five market vendors were selected at each municipal market as well as ten individual roadside stalls. In total 35 vendors were surveyed. From each vendor, a sample of at least 10 fruits, for each cultivar available, was purchased for the purpose of postharvest disease assessment in the laboratory. Seven mango cultivars were collected in total during the survey – these included the five local cultivars studied during the field survey, as well as 'Parrot' and 'Kensington Pride'.

The fruit samples were placed in fruit boxes, stored at 23°C and were assessed for postharvest disease (body and stem end rots) when the majority of fruits in the sample of 10 fruits were at the eating ripe stage (firmness stage 4). The incidence of body and stem end rots caused by *Colletotrichum* spp., and the incidence of body and stem end rots caused by Botryosphaeriaceous fungi, were recorded separately for each sample. Different symptom types are shown in Figure 1.

Fungal isolations and preliminary identifications

For both the field and market surveys, fungal isolations were conducted on fruits showing anthracnose or stem end rot symptoms at the end of the assessment period. Isolations were also conducted on a small sample of 17 fruits (seven 'Carabao' and ten 'Kerosene' fruits) not directly included in the surveys reported here.

Fruits were surface sterilised by wiping with 70% (v/v) ethanol before excising four tissue pieces from lesion margins aseptically using a sterile scalpel blade. Tissue pieces were then transferred onto ½ strength potato dextrose agar (PDA -20g Difco™ potato dextrose agar powder) plates, which were incubated at 25°C. To obtain pure cultures, colonies were subcultured after 2–3 days by aseptically removing a 5mm square piece of agar + fungus from the edge of the colony and transferring it onto the centre of a fresh ½ strength PDA plate. Colonies were hyphal tipped after a further 2–3-day period, and the resultant colonies were incubated at 25°C under 12 h/day irradiation from near-ultraviolet light (310–410 nm UV-A) and 12 h/day darkness to promote sporulation. Cultures were then stored in the Plant Pathology Culture Collection at Koronivia Research Station as small pieces (approx. 5 mm3) of agar + fungus in tubes of sterile distilled water. A duplicate set of cultures (in tubes of sterile distilled water) were prepared for all isolates identified as *Colletotrichum* sp. based on morphological characteristics (colony appearance and microscopic examination of conidia). This set of cultures was imported under permit into Australia for molecular species identification.

Molecular identification of fungal isolates

The *Colletotrichum* isolates collected during the 2015/16 and 2016/17 mango seasons in Fiji (from field and market surveys) were imported into Australia for molecular identification under DAWR Import Permit no. IPI 5000786 and were held in the QC3

Figure 1. Symptoms of postharvest diseases in mango: a) anthracnose lesions caused by *Colletotrichum* sp. on the fruit body; b) anthracnose lesion caused by *Colletotrichum* sp. at the fruit stem end; c) anthracnose lesions caused by *Colletotrichum* sp. on the body and stem end of fruits at a Fijian fruit market; d) lesions caused by a Botryosphaeriaceous fungal species on the body of fruits at a Fijian fruit market; e) lesion caused by a Botryosphaeriaceous fungal species at the fruit stem end.

laboratory at the Ecosciences Precinct (DAF) in Brisbane, where DNA extractions were undertaken.

Live cultures imported from Fiji were subcultured on fresh ½ strength PDA plates amended with streptomycin in the QC3 laboratory. Cultures were also stored in 15%

glycerol and maintained at −80°C in the same laboratory. Fungal hyphae were scraped from 10-day old cultures with a sterile plastic loop and macerated with 0.5 mm glass beads (Daintree Scientific) in a Tissue Lyser (QIAGEN). Genomic DNA was extracted using the BioSprint 15 (QIAGEN) according to the manufacturer's instructions. The primers used to amplify the ITS (internal transcribed spacer), TUB (β-tubulin 2) and CAL (calmodulin) gene regions of the fungal DNA extracts are shown in Table 1. Phusion High Fidelity PCR Master Mix (New England Biolab) was used to amplify all gene regions. Purification and sequencing of PCR products was done by Macrogen Inc. (South Korea) on the 3730xl DNA Analyser (Applied Biosystems) using amplifying primers. Geneious software v.9.1 (Biomatters Ltd.) was used to assemble all sequences that were generated. Comparisons were made against type specimens by conducting BLAST searches (https://blast.ncbi.nlm.nih.gov/Blast.cgi).

All isolates of *Colletotrichum* spp. obtained from field and market surveys (126 isolates in total) were first sequenced using ITS primers to gain a preliminary identification. When an identification based on ITS was the species *asianum* (122 isolates), in the most part no further sequencing was undertaken, as ITS is known to distinguish *C. asianum* from other *Colletotrichum* species reliably (Weir et al. 2012). However, further sequencing based on the TUB and CAL gene regions was performed on a subset of these isolates (30 isolates) to confirm an identification of *C. asianum*. Only four of the 126 isolates were identified as a species other than *asianum* at the ITS region. These were sequenced at the TUB and CAL gene regions to confirm species identity.

As there was no permit for the import of live cultures of fungi from the Botryosphaeriaceae into Australia, DNA extractions of the stem end rot fungi isolated from mangoes in the field and market surveys were performed in the laboratories of the Koronivia Research Station and the Secretariat of the Pacific Community (SPC). Total genomic DNA was extracted from fungal hyphae scraped from 8–10 days old cultures growing on ½ strength PDA plates, macerated and extracted using the ISOLATE II Plant DNA kit (Bioline) according to the manufacturers' instructions. The DNA product was then imported (DAWR Import Permit no. 0000753489 for Nucleic acids) into Australia for molecular identification and held in the QC3 laboratory, DAF, Ecosciences Precinct in Brisbane.

Table 1. Primers used in the identification of fungi isolated from mango fruits in Fiji.

Gene	Product name	Primer	Direction	Sequence (5'– 3')	Reference
ITS	Internal transcribed	V9G	F[1]	TTACGTCCCTGCCCTTTGTA	de Hoog and Gerrits van den Ende (1998)
	spacer	ITS4	R	TCCTCCGCTTATTGATATGC	White et al. (1990)
TUB	ß-tubulin 2	T1	F	AACATGCGTGAGATTGTAAGT	O'Donnell and Cigelnik (1997)
		Bt2b	R	ACCCTCAGTGTAGTGACCCTTGGC	Glass and Donaldson (1995)
CAL	Calmodulin	CL1C	F	GAATTCAAGGAGGCCTTCTC	Weir et al. (2012)
		CL2C	R	CTTCTGCATCATGAGCTGGAC	Weir et al. (2012)
TEF-1α	Translation elongation factor 1	EF1-1018	F	GAYTTCATCAAGAACATGAT	Stielow et al. (2015)
		EF1-1620	R	GACGTTGAADCCRACRTTGTC	Stielow et al. (2015)
		EF1-728F	F	CATCGAGAAGTTCGAGAAGG	Carbone and Kohn (1999)
		EF2	R	GGARGTACCAGTSATCATGTT	O'Donnell et al. (1998)
		EF1	F	ATGGGTAAGGARGACAAGAC	O'Donnell et al. (1998)

[1]F = forward; R = reverse

The primers used to amplify the ITS (internal transcribed spacer) and TEF-1α (translation elongation factor 1) gene regions of the DNA extracts from stem end rot fungal isolates are shown in Table 1. A range of primers were tested for sequencing at the TEF region. Best results were obtained when the EF1-728F and EF2 primer pair was used. PCR and sequencing were done as previously described. Sequencing results were sometimes equivocal and non-type BLAST searches were used as well as type searches to clarify identity for some isolates.

Data analysis

All statistical analyses were conducted using Genstat (2022). Models were initially fitted with the treatment terms as well as their interactions, and non-significant interactions were subsequently dropped from models. Residual plots were assessed for deviation from model assumptions. Means and standard errors were calculated along with pairwise comparisons for significant treatment terms. The pairwise comparisons were performed using Fisher's Protected Least Significant Difference (LSD) at a significance level of 5%, where means with the same letter are not significantly different. For the field survey, analyses of stem end and body rot at eating ripe were performed using an ANOVA. Treatment effects were cultivar, locality and their interaction for the 2015/16 study (local cultivars at five localities), and cultivar for the 2016/17 study (imported cultivars at one locality) with tree and rep as the random effects respectively. For the market survey, disease incidence was analysed using a Generalised Linear Model (GLM) with a binomial distribution and logit link. Cultivar, locality, and their interaction were included as treatment effects.

Results

Field survey of local cultivars

The severity of body rot in fruits sampled for the field survey of local mango cultivars varied considerably according to cultivar and location (Figure 2). At most localities, 'Maqo Loa' fruits had the highest severity of body rot at the eating ripe stage, compared to other cultivars. 'Salusalu' and 'Maqo Uto' on average had the lowest severity of body rots (3 and 6% respectively). Notable exceptions were high levels of body rots in 'Maqo Uto' fruits from Tavua, and in 'Salusalu' fruits from Nadi. For locality, fruits from Nadi on average had the highest severity of body rots at the eating ripe stage (16%), whereas fruits from Ba on average had the lowest (4%).

Variable stem end rot levels were also found in the field survey of local mango cultivars and growing localities (Figure 3). 'Peach' mango fruits on average had the highest severity of stem end rot at the eating ripe stage (12%) compared to fruits of the other cultivars, whereas 'Salusalu' had on average the lowest severity (2%). For locality, stem end rot severity ranged from an average of 10% for Nadi and Lautoka to only 3% for Ba.

Figure 4 shows the skin colour ratings of eating ripe fruits harvested from the five local cultivars at each of five different localities. While there was a significant interaction between cultivar and locality, 'Maqo Loa' fruits in most instances had lower skin
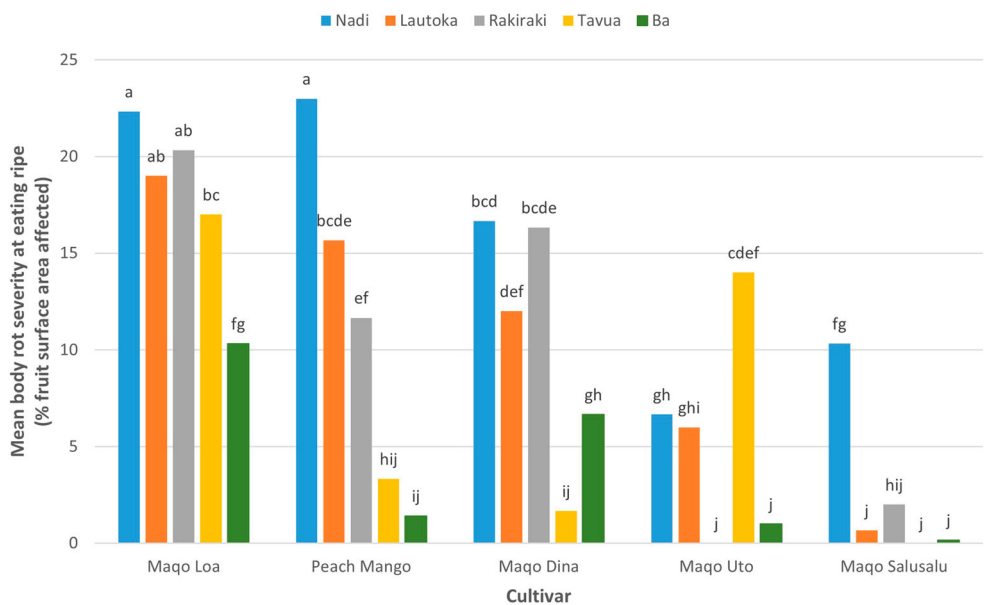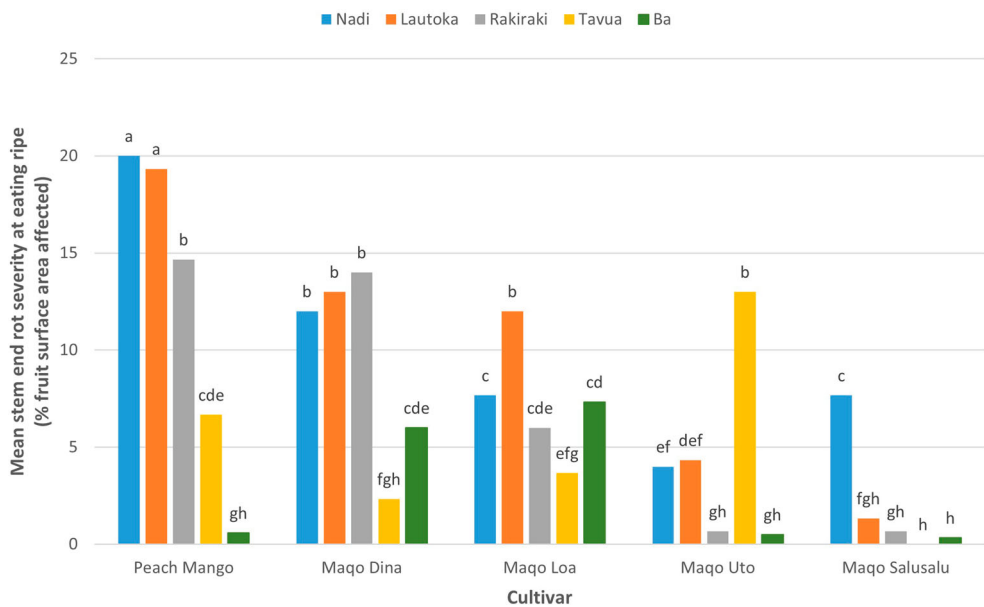
Figure 2. Field survey – the severity of body rot (at the eating ripe stage) of fruits harvested from five local mango cultivars at each of five different localities in Fiji. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.

colour ratings at the eating ripe stage than the other cultivars. Overall, 'Salusalu' and 'Maqo Dina' fruits had the highest skin colour ratings, although there were some exceptions, particularly in the case of fruits from Nadi.

Figure 3. Field survey – the severity of stem end rot (at the eating ripe stage) of fruits harvested from five local mango cultivars from each of five different localities in Fiji. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.
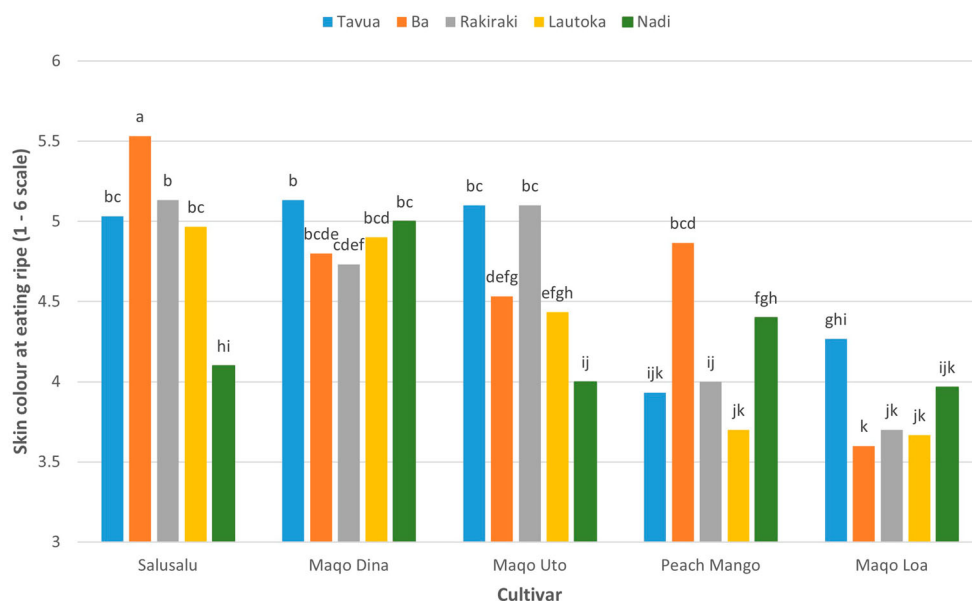
Figure 4. Field survey – skin colour rating (at the eating ripe stage) of fruits harvested from five local mango cultivars from each of five different localities in Fiji. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.

Fruit shelf-life also varied significantly with cultivar and locality (Figure 5), with 'Maqo Loa' fruits having on average the longest shelf-life (9.8 days to eating ripe), and 'Maqo Uto' and 'Maqo Dina' having the shortest (6.9 and 7.2 days respectively).

Field survey of imported cultivars

In the field survey of imported cultivars, 'Nam Doc Mai' had the lowest severity of body rot at eating ripe, whereas 'Tahitian' and the local cultivar 'Maqo Dina' had the highest (Figure 6). 'Nam Doc Mai' also had the lowest severity of stem-end rot of all cultivars assessed, while 'Nang Klang Wan' had the highest (Figure 7). 'Nang Klang Wan' fruits also had significantly higher skin colour ratings at eating ripe than fruits from all cultivars except for 'Nam Doc Mai' (Figure 8). 'Tommy Atkins' and 'Tahitian Mango' fruits had the lowest skin colour ratings at eating ripe. Fruit shelf-life varied from 10.6 days in 'Kensington Pride' to 5.4 days in 'Nam Doc Mai' (Figure 9).

Market survey of mango postharvest diseases in Fiji

The most common cultivars available at markets overall were 'Maqo Dina' and 'Maqo Uto', while 'Kensington Pride' was only available at one roadside stall (Figure 10). Lesions caused by *Colletotrichum* spp. were found on both the body and the stem end of the fruits (Figure 1). While lesions caused by Botryosphaeriaceous fungi were predominantly found on the stem end of fruits, some did occur on the body of fruits.

'Parrot' and 'Kensington Pride' fruits had a 100% incidence of *Colletotrichum* body rot, which was significantly higher than in all other cultivars (Figure 11). 'Salusalu'
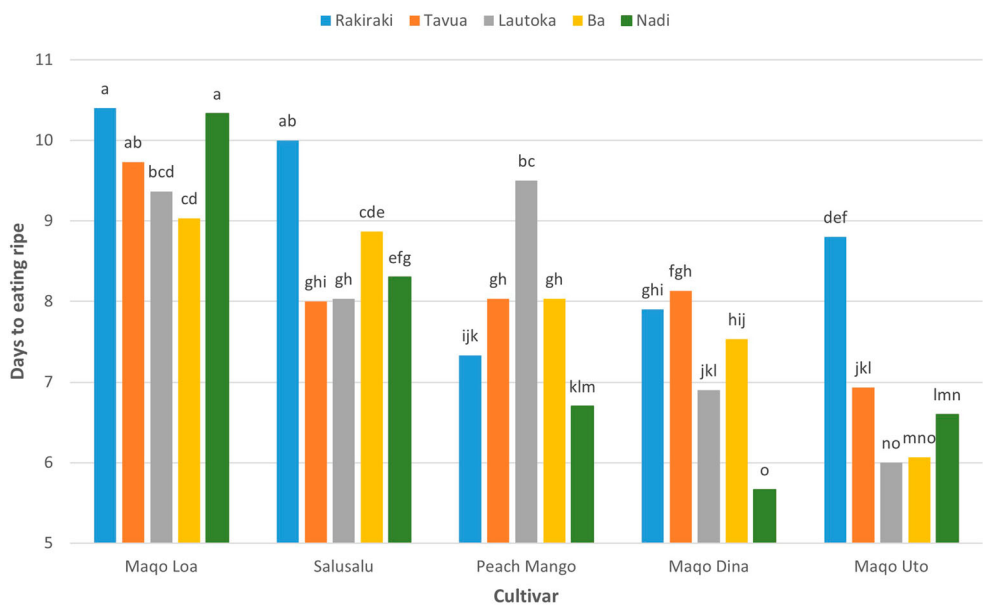
Figure 5. Field survey – shelf-life (days to eating ripe) of fruits harvested from five local mango cultivars from each of five different localities in Fiji. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.
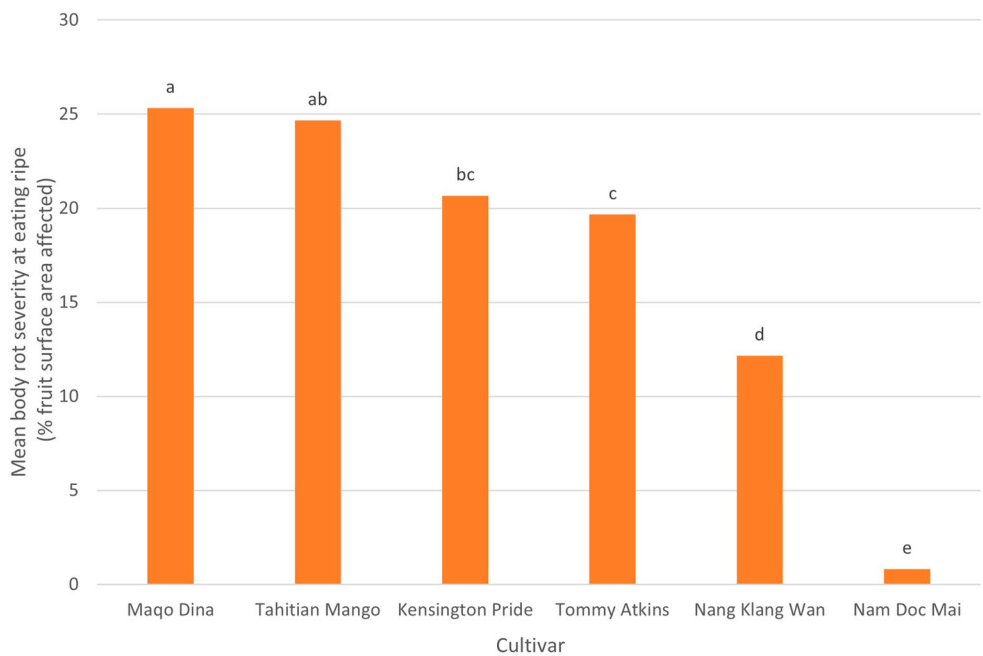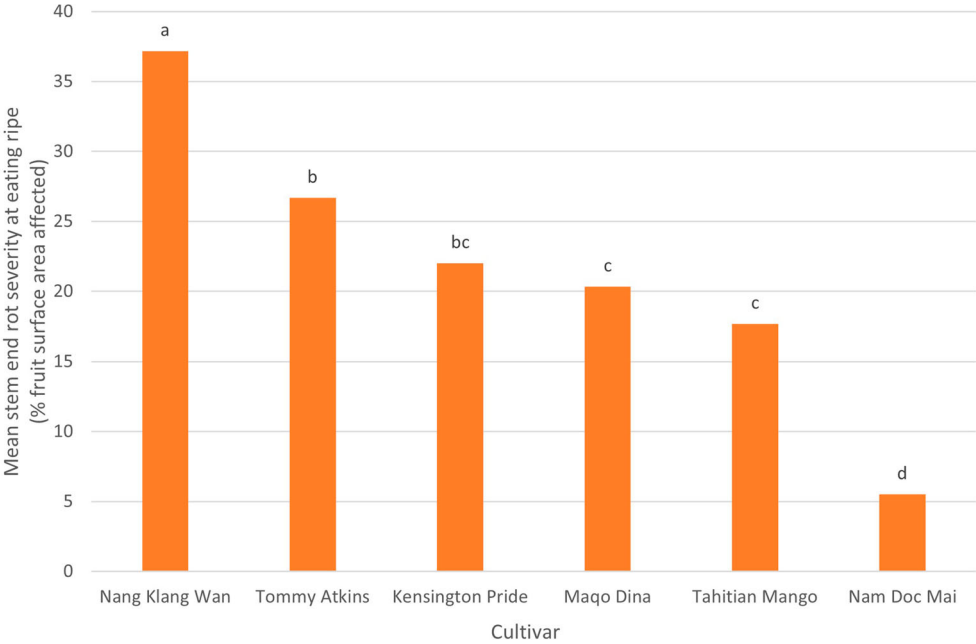

Figure 6. Field survey – the severity of body rot (at the eating ripe stage) in mango fruits harvested from five imported cultivars and one local cultivar in the Nadi area. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.
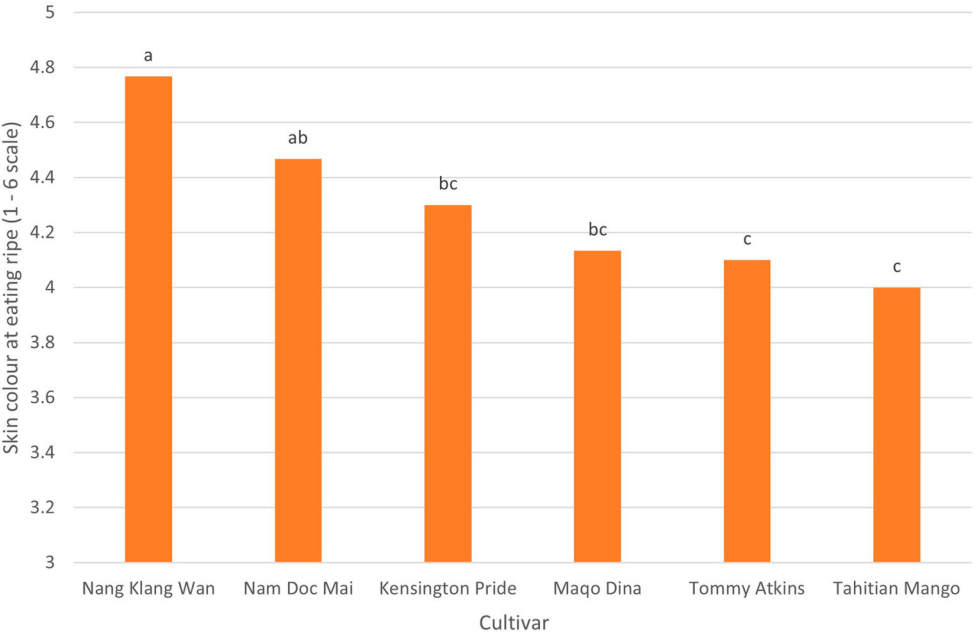
Figure 7. Field survey – the severity of stem end rot (at the eating ripe stage) in mango fruits harvested from five imported cultivars and one local cultivar in the Nadi area. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.

Figure 8. Field survey – skin colour rating (at the eating ripe stage) in mango fruits harvested from five imported cultivars and one local cultivar in the Nadi area. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.
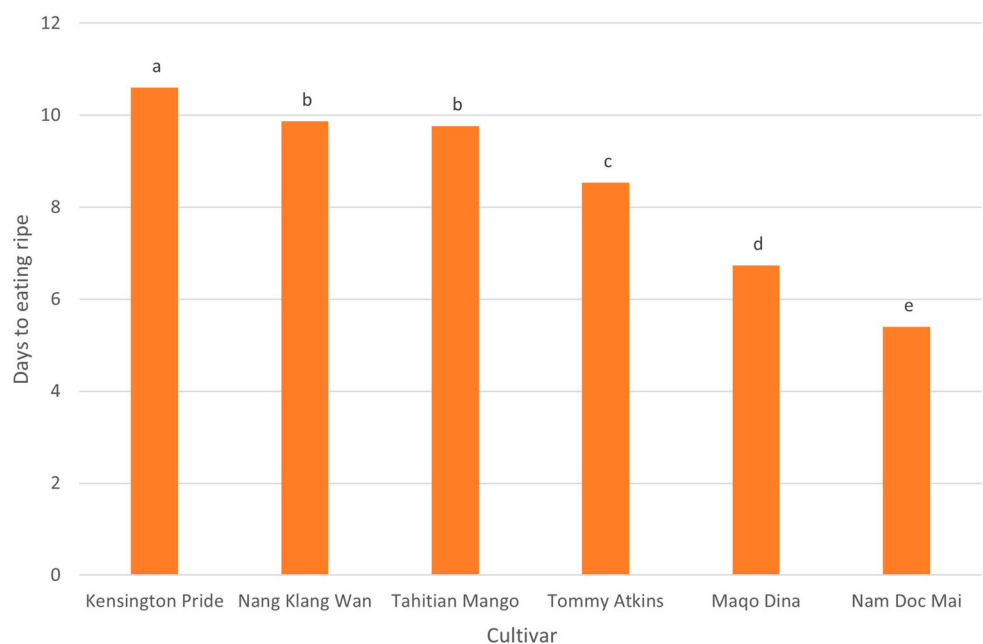
Figure 9. Field survey – shelf-life (days to eating ripe) of fruits harvested from five imported cultivars and one local cultivar in the Nadi area. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.
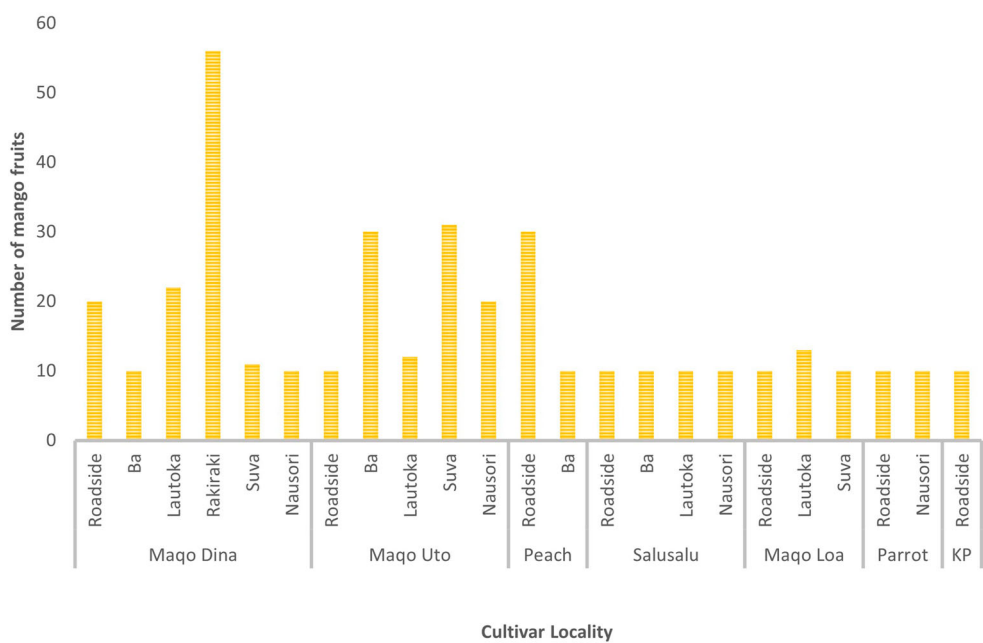

Figure 10. Market survey – total fruit numbers collected for each mango cultivar at each market locality in Fiji. *KP* = Kensington Pride.
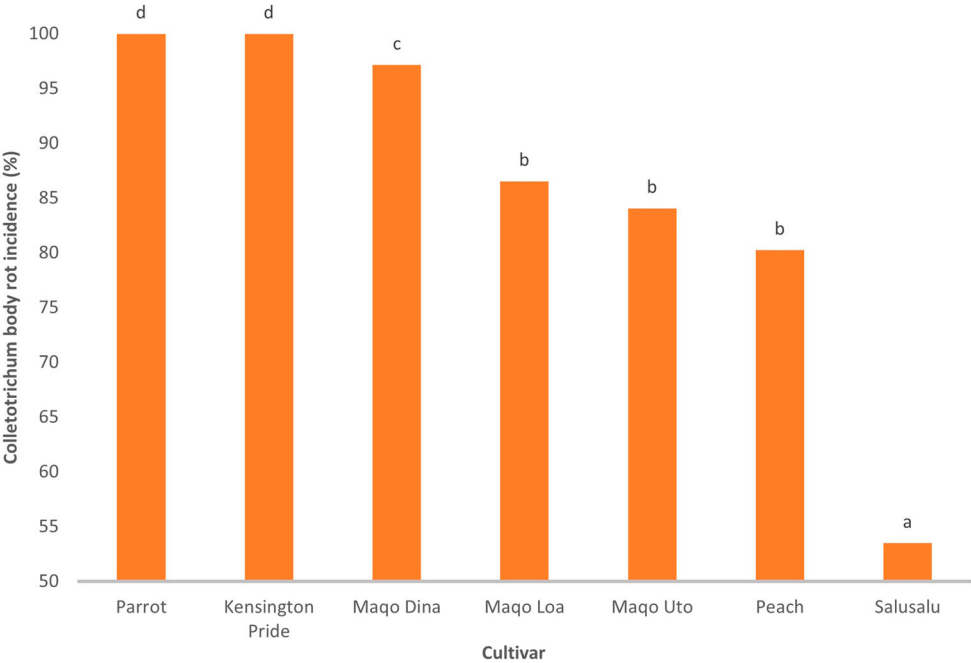
Figure 11. Market survey – the incidence of body rot caused by *Colletotrichum* spp. in mango fruits of different cultivars (averaged over all markets). Means surmounted by the same letter are not significantly different using the LSD test at *P* = 0.05.
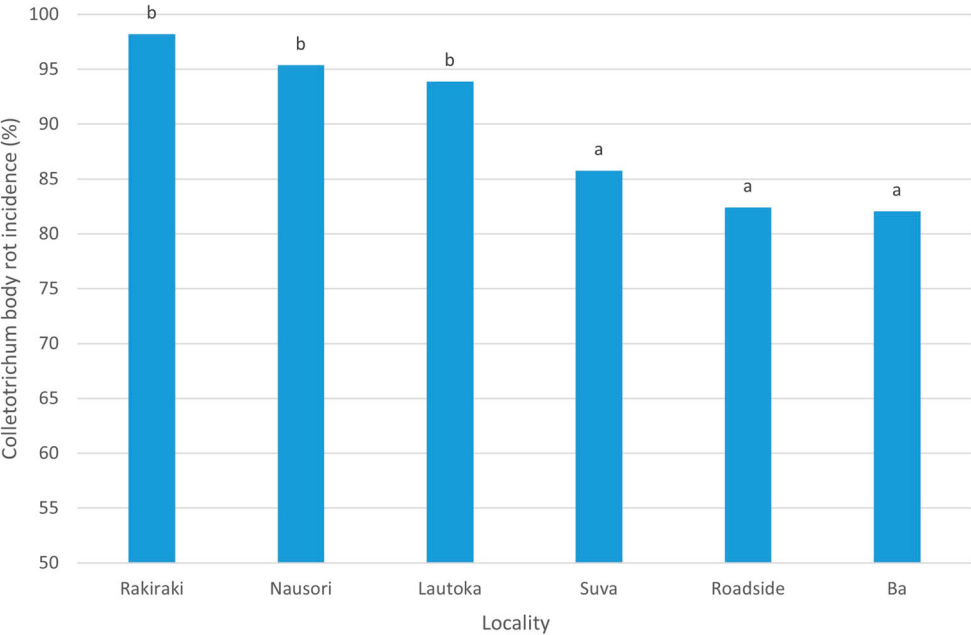

Figure 12. Market survey – the incidence of body rot caused by *Colletotrichum* spp. in mango fruits purchased from different municipal markets and roadside stalls in Fiji (averaged over all cultivars). Means surmounted by the same letter are not significantly different using the LSD test at *P* = 0.05.

fruits had approximately half the incidence of *Colletotrichum* body rot compared to 'Parrot' and 'Kensington Pride'. In terms of locality, fruits collected from markets in Rakiraki, Nausori and Lautoka had significantly higher incidence levels for *Colletotrichum* body rot than those from Suva, Ba and the roadside stalls (Figure 12).

For stem end rot caused by *Colletotrichum* spp., there was a significant interaction between cultivar and market locality (Figure 13). 'Maqo Dina' fruits from Suva market and 'Salusalu' fruits from Lautoka market had the highest incidence of *Colletotrichum* stem end rot of all the fruits collected in the survey. However, when averaged over all markets, 'Maqo Loa' and 'Maqo Dina' fruits had the highest incidence of *Colletotrichum* stem end rot (33%), while 'Parrot' and 'Kensington Pride' fruits had none (noting that these were smaller sample sizes than the other cultivars). In terms of locality, fruits from Suva and Rakiraki markets had the highest incidence of *Colletotrichum* stem end rot on average (64 and 63% respectively), with Ba and Nausori having the lowest incidence on average (2.2 and 0% respectively).

Recorded levels of body rot caused by Botryosphaeriaceous fungi were relatively low in the market survey, however differences were seen at the different localities where samples were taken (Figure 14). Fruits from Lautoka markets and the roadside stalls had a significantly higher incidence of Botryosphaeriaceous body rot than those from Suva, Rakiraki and Nausori markets, with the latter two markets having no Botryosphaeriaceous body rot at all. There were no significant effects of cultivar (data not shown).
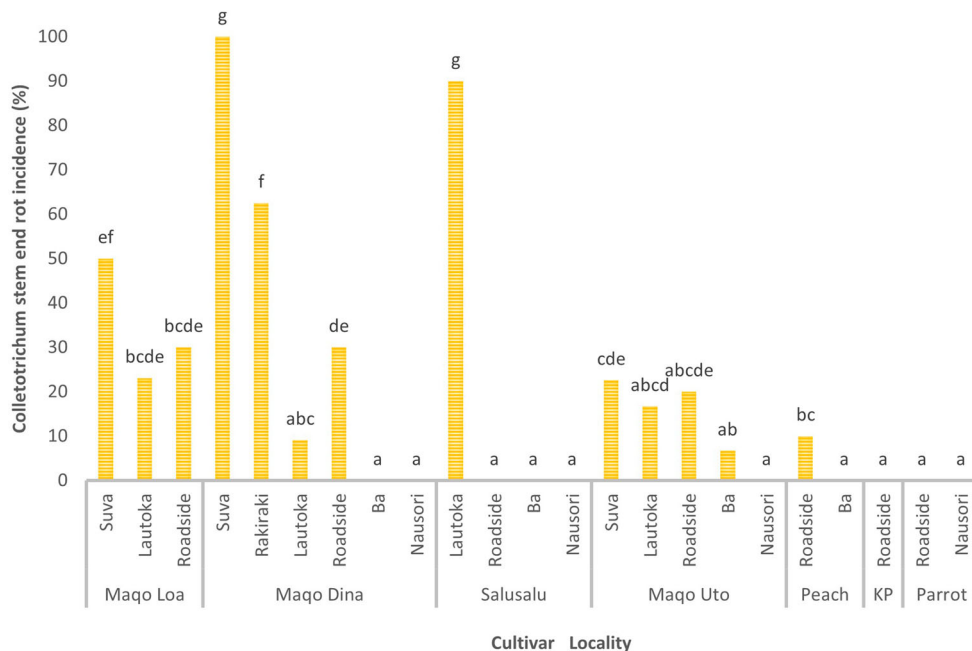

Figure 13. Market survey – the incidence of lesions caused by *Colletotrichum* spp. at the stem-end of mango fruits of different cultivars purchased from different municipal markets and roadside stalls in Fiji.
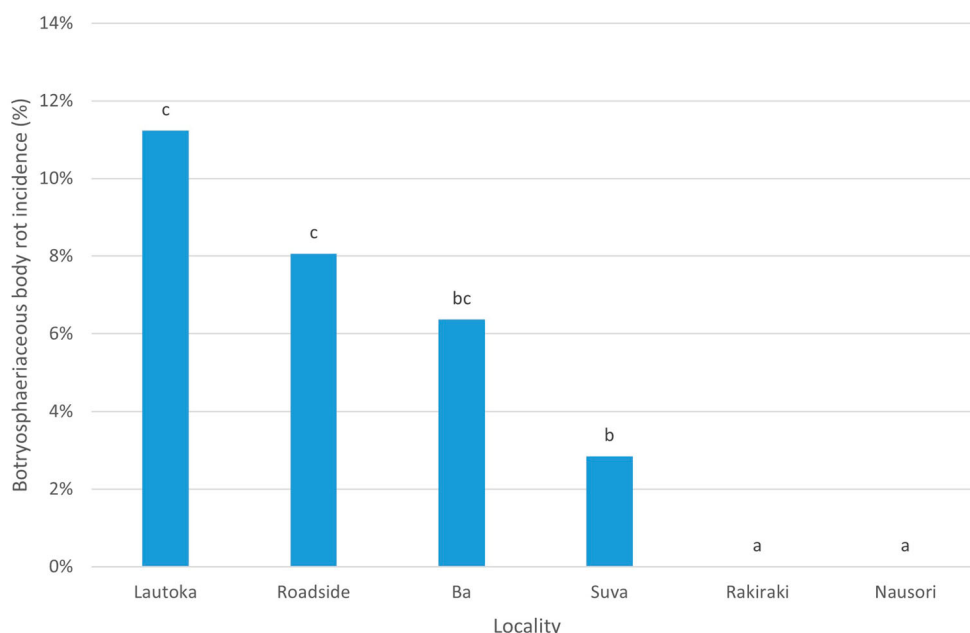
Figure 14. Market survey – the incidence of body rot caused by Botryosphaeriaceous fungi in mango fruits purchased from different municipal markets and roadside stalls in Fiji (averaged over all cultivars). Means surmounted by the same letter are not significantly different using the LSD test at *P* = 0.05.

The incidence of Botryosphaeriaceous stem end rot did however vary with cultivar and locality in the market survey (Figure 15). On average, 'Parrot' and 'Salusalu' fruits had higher incidence values (77 and 58% respectively) than other cultivars. Fruits from the Nausori market had on average more Botryosphaeriaceous stem end rot (83%) than those from the other markets.

Molecular identification of fungal isolates

Isolates of *Colletotrichum* spp. were collected from local and imported mango cultivars as part of the field and market surveys. A total of 126 *Colletotrichum* isolates were isolated during the study and imported into Australia under permit for molecular species identification. All isolates were initially sequenced at the ITS gene region. Of the 126 isolates sequenced; 122 isolates (96.8%) were identified as *C. asianum* using ITS primers. While the ITS region is known to reliably differentiate *C. asianum* from other *Colletotrichum* species (Weir et al. 2012), a subset of 30 putative *C. asianum* isolates sequenced at the TUB and CAL regions confirmed their identity.

While *C. asianum* was by far the predominant fungus isolated from anthracnose lesions in Fiji, four isolates out of the total of 126 isolates were identified as other species – two isolates of *C. simmondsii* and two isolates of *C. fructicola* (both identified by sequencing at the ITS, TUB and CAL regions).

A total of 32 fungal isolates were cultured from stem end rot lesions as part of the field and market surveys and imported into Australia under permit. Using both ITS and TEF
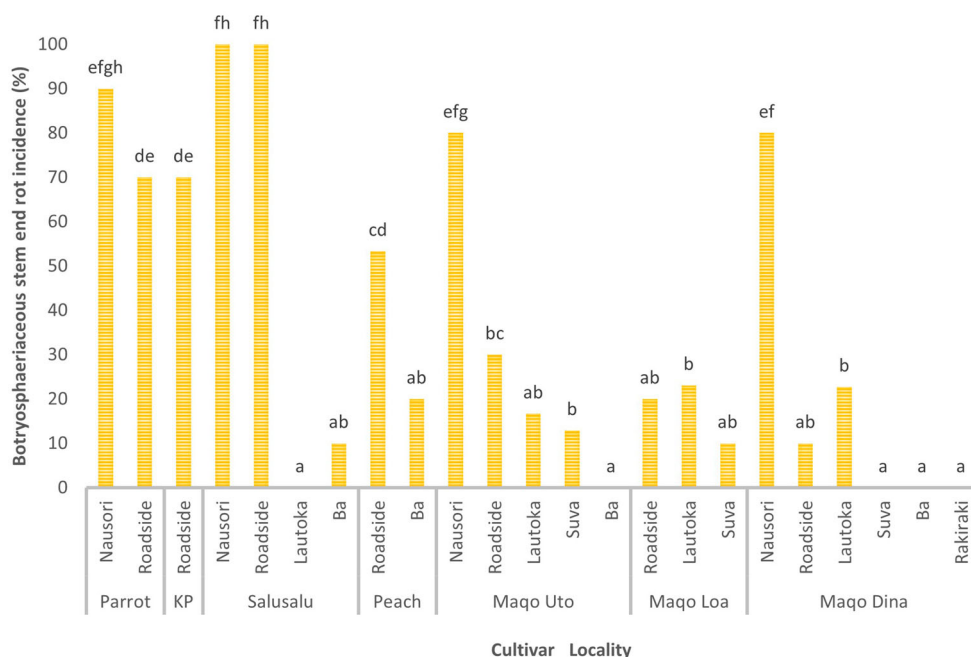
Figure 15. Market survey – the incidence of stem end rot caused by Botryosphaeriaceous fungi in mango fruits of different cultivars purchased from different municipal markets and roadside stalls in Fiji. KP = Kensington Pride. Means surmounted by the same letter are not significantly different using the LSD test at $P = 0.05$.

primers (see Table 1 for primers used), 11 isolates were identified as *Neofusicoccum parvum*, nine as *Lasiodiplodia theobromae*, three as *L. brasiliensis,* one as *N. umdonicola* and one as *N. kwambonambiense*. An additional five isolates were tentatively identified as *L. theobromae* on the basis of ITS only (3 isolates) or TEF only (2 isolates). One isolate was identified as *Diaporthe lithicola* using ITS primers but gave an inconclusive result using TEF primers. One isolate of *Neofusicoccum* sp. could not be resolved at the species level.

Discussion

Collective results from the field and the market surveys confirm that anthracnose (predominantly caused by *Colletotrichum asianum*) and stem end rot (predominantly caused by *Lasiodiplodia theobromae* and *Neofusicoccum parvum*) are highly prevalent postharvest diseases affecting mango fruits in Fiji. While this may not be surprising given the ubiquitous nature of these diseases on mango worldwide, the levels of their occurrence in mangoes in Fiji were extremely high, with some cultivars such as 'Maqo Dina' showing anthracnose incidence levels of 100%. Given 'Maqo Dina' is the most widely planted mango cultivar in Fiji (Iqbal 1982), this would translate to very high postharvest fruit losses across the mango industry.

Levels of stem end rot caused by Botryosphaeriaceous fungi were higher than expected in markets, with some cultivars such as 'Parrot' showing a stem end rot incidence of

nearly 80%. In Australia postharvest losses of up to 20% of the harvested mango crop can typically be attributed to stem end rot (Plan et al. 2002), although Iram et al. (2014) reported up to 80% losses in Pakistan. Due to the lack of access to quality disease-free fruits, Fijian consumers have become tolerant of mangoes affected by postharvest disease, typically cutting out areas of affected flesh before consuming the unaffected portions (M. Lomavatu, personal observation). Future development of the mango industry in Fiji, even for domestic consumption, will require implementation of appropriate and improved integrated disease management strategies.

In the field survey of local cultivars, the incidence and severity of body rots was consistently lowest in the cultivar 'Salusalu'. This finding was also supported by the results of the market survey. 'Salusalu' is the only mango cultivar that can be grown successfully in the Suva region (Baker 2015), which is one of the wettest mango production areas of Viti Levu. 'Salusalu' also attained one of the highest skin colour ratings at eating ripe in this study (average colour rating of 5 in field survey) and had an acceptable shelf-life (average of 8.6 days to eating ripe in field survey). Although generally considered as a 'local' cultivar in Fiji, 'Salusalu' is likely to have South African origins (Baker 2015). 'Nam Doc Mai' fruits also showed remarkably low body rot severity at eating ripe in the field survey of imported cultivars, as well as low stem end rot severity and good skin colour development, but with a shorter shelf-life (average of 5.4 days to eating ripe in field survey) compared to 'Salusalu'.

In terms of anthracnose causal agents in Fiji, approximately 97% of the *Colletotrichum* isolates obtained were identified as *C. asianum*, which belongs within the *C. gloeosporioides* species complex (Weir et al. 2012). This is consistent with reports from other countries such as Australia, Thailand, The Philippines, Brazil, and Panama where *C. asianum* has also been found to be an important causal agent of anthracnose in mango (Phoulivong et al. 2010; Weir et al. 2012; Lima et al. 2013; Shivas et al. 2016; Mitchell et al. 2017; Giblin et al. 2018). The pathogenicity of *C. asianum* on detached mango fruit (cv. Dina) in Fiji was demonstrated in Lomavatu (2019).

The two other species identified in association with mango anthracnose symptoms in Fiji were *C. simmondsii* and *C. fructicola. C. simmondsii* belongs within the *C. acutatum* species complex (Weir et al. 2012), which is consistent with an older report which cites *C. acutatum* as a minor pathogen causing anthracnose in mango in Australia (Fitzell 1979). Like *C. asianum, C. fructicola* belongs within the *C. gloeosporioides* species complex (Weir et al. 2012), and has been reported in association with mango anthracnose in Brazil (Lima et al. 2013) and Australia (Mitchell et al. 2017). All three species (*C. asianum, C. fructicola* and *C. simmondsii*) represent first reports for Fiji.

Lasiodiplodia theobromae and *Neofusicoccum parvum* were the most common species found in association with mango stem end rot symptoms in Fiji. These two species have been reported as the major causal agents of stem end rot in mango worldwide (Johnson et al. 2011). *L. theobromae* is usually more common in tropical regions, whereas *N. parvum* usually predominates in subtropical environments. The pathogenicity of *L. theobromae* and *N. parvum* on detached mango fruit (cv. Dina) in Fiji was demonstrated in Lomavatu (2019).

Other species from the Botryosphaeriaceae isolated from mango stem end rot symptoms in Fiji were identified as *Lasiodiplodia brasiliensis*, *Neofusicoccum umdonicola* and *N. kwambonambiense*. Each of these species have been reported to occur in association

with mango in other countries (Coutinho et al. 2017; Mehl et al. 2017; Tan et al. 2019). These species represent new reports for Fiji.

Acknowledgments

Disclosure statement

No potential conflict of interest was reported by the author(s).

Funding

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References

Abdollahzadeh J, Mohammadi GE, Javadi A, Shams-bakhsh M, Zare R, Phillips AJL. 2010. Phylogeny and morphology of four new species of *Lasiodiplodia* from Iran. Persoonia. 23:1–8. doi:10.3767/003158509X467552.

Arauz LF. 2000. Mango anthracnose: economic impact and current options for integrated management. Plant Disease. 84:600–611. doi:10.1094/PDIS.2000.84.6.600.

Baker I. 2015. Assess current state of mango export industry in Fiji. Australian Centre for International Agricultural Research, 36 pages.

Brown MF. 2010. Value chain study of Tropical Dried Fruits in Fiji - Promoting Healthy Snacks. Facilitating Agriculture Commodity Trade in the Pacific [FACT].

Carbone I, Kohn LM. 1999. A method for designing primer sets for speciation studies in filamentous ascomycetes. Mycologia. 91:553–556. doi:10.1080/00275514.1999.12061051.

Coutinho IBL, Freire FCO, Lima CS, Lima JS, Goncalvesc FJT, Machado AR, Silvae AMS, Cardosob JE. 2017. Diversity of genus *Lasiodiplodia* associated with perennial tropical fruit plants in northeastern Brazil. Plant Pathology. 66:90–104. doi:10.1111/ppa.12565.

de Hoog GS, Gerrits van den Ende AHG. 1998. Molecular diagnostics of clinical strains of filamentous Basidiomycetes. Mycoses. 41:183–189. doi:10.1111/j.1439-0507.1998.tb00321.x.

FAOSTAT. 2023. FAO Statistics, Food and agriculture organization of the United Nations. Rome: FAO. http://faostat.fao.org/.

Fitzell RD. 1979. *Colletotrichum acutatum* as a cause of anthracnose of mango in New South Wales. Plant Disease Reporter. 63:1067–1070.

Genstat. 2022. Genstat for Windows, release 22.1. Oxford: VSN International Ltd.

Giblin FR, Tan YP, Mitchell R, Coates LM, Irwin JAG, Shivas RG. 2018. *Colletotrichum* species associated with pre-and post-harvest diseases of avocado and mango in eastern Australia. Australasian Plant Pathology. 47:269–276. doi:10.1007/s13313-018-0553-0.

Glass NL, Donaldson GC. 1995. Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. Applied and Environmental Microbiology. 61:1323–1330. doi:10.1128/aem.61.4.1323-1330.1995.

Hassan MK. 2006. Constitutive alk(en)ylresorcinols and resistance to postharvest diseases in mango (*Mangifera indica* L.). PhD thesis, School of Agronomy and Horticulture. University of Queensland Brisbane, Australia, 286 pp.

Honger JO, Offei SK, Oduro KA, Odamtten GT, Nyaku ST. 2014. Identification and species status of the mango biotype of *Colletotrichum gloeosporioides* in Ghana. European Journal of Plant Pathology. 140:455–467. doi:10.1007/s10658-014-0480-z.

Iqbal M. 1982. Review of mango research and production in Fiji. Fiji Agricultural Journal. 44:21–26.

Iram S, Rasool A, Ahmad I. 2014. Comparison of incidence, prevalence and severity of postharvest fungal diseases in Pakistan improved integrated management orchards and conventional practices blocks. International Journal of Scientific & Engineering Research. 5:1274–1284.

Ismail AM, Cirvilleri G, Polizzi G, Crous PW, Groenewald JZ, Lombard L. 2012. *Lasiodiplodia* species associated with dieback disease of mango (*Mangifera indica*) in Egypt. Australasian Plant Pathology. 41:649–660. doi:10.1007/s13313-012-0163-1.

Johnson GI. 1994. Stem end rot. In: Ploetz R.C., Zentmyer G.A., Nishijima W.T., Rohrbach K.G., Ohr H.D., editors. Compendium of tropical fruit diseases. St Paul, Minnesota: APS Press; p. 39–41.

Johnson GI. 2008. Literature Review to determine the options and solutions to postharvest disease management for the mango industry. Horticulture Australia Limited, Final Report for project MG08017, Horticulture 4 Development, Canberra, 132 p.

Johnson GI, Coates LM, Cooke AW. 2011. Status of Mango post-harvest disease management R&D: Options and solutions for the Australian Mango Industry. International Workshop on Diagnosis & Control of Postharvest Diseases Berrimah Research Station, Darwin, Australia.

Li L, Mohd MH, Mohamed Nor NMI, Subramaniam S, Latiffah Z. 2021. Identification of Botryosphaeriaceae associated with stem-end rot of mango (*Mangifera indica* L.) in Malaysia. Journal of Applied Microbiology. 130:1273–1284. doi:10.1111/jam.14828.

Lima NB, de A, Batista MV, De Morais MA, Barbosa MAG, Michereff SJ, Hyde KD, Câmara MPS. 2013. Five *Colletotrichum* species are responsible for mango anthracnose in northeastern Brazil. Fungal Diversity. 61:75–88. doi:10.1007/s13225-013-0237-6.

Liu LP, Shu J, Zhang L, Hu R, Chen CQ, Yang LN, Lu BH, Liu YN, Yu L, Wang X, et al. 2017. First report of post-harvest anthracnose on mango (*Mangifera indica*) caused by *Colletotrichum siamense* in China. Plant Disease. 101:833–833. doi:10.1094/PDIS-08-16-1130-PDN.

Lomavatu MF. 2019. Postharvest Diseases of Mango in Fiji. PhD thesis, University of the Sunshine Coast, February 2019.

Marques MW, Lima NB, De MoraisJrMA, Barbosa MAG, Souza BO, Michereff SJ, Phillips AJL, Câmara MPS. 2013. Species of *Lasiodiplodia* associated with mango in Brazil. Fungal Diversity. 61:181–193. doi:10.1007/s13225-013-0231-z.

Mehl JWM, Slippers B, Roux J, Wingfield MJ. 2017. Overlap of latent pathogens in the Botryosphaeriaceae on a native and agricultural host. Fungal Biology. 121:405–419. doi:10.1016/j.funbio.2016.07.015.

Mitchell RW, Coates LM, Tan YP, Dann EK, Dean JR, Giblin FR, Anderson JM, Shivas RG. 2017. *Colletotrichum* species associated with anthracnose of subtropical and tropical fruit in Australia, Science Protecting Plant Health Conference, Poster Presentation, Brisbane.

Ni HF, Yang HR, Chen RS, Liou RF, Hung TH. 2012. New *Botryosphaeriaceae* fruit rot of mango in Taiwan: identification and pathogenicity. Botanical Studies. 53:467.

O'Donnell K, Cigelnik E. 1997. Two divergent intragenomic rDNA ITS2 types within a monophyletic lineage of the fungus *Fusarium* are nonorthologous. Molecular Phylogenetics and Evolution. 7:103–116. doi:10.1006/mpev.1996.0376.

O'Donnell K, Kistler HC, Cigelnik E, et al. 1998. Multiple evolutionary origins of the fungus causing Panama disease of banana: Concordant evidence from nuclear and mitochondrial

gene genealogies. Proceedings of the National Academy of Sciences of the United States of America. 95:2044–2049. doi:10.1073/pnas.95.5.2044.

Pavlic D, Wingfield MJ, Barber P, Slippers B, Hardy GESJ, Burgess TI. 2008. Seven new species of the Botryosphaeriaceae from baobab and other native trees in Western Australia. Mycologia. 100:851–866. doi:10.3852/08-020.

Phillips AJL, Alves A, Abdollahzadeh J, Slippers B, Wingfield MJ, Groenewald JZ, Crous PW. 2013. The *Botryosphaeriaceae*: genera and species known from culture. Studies in Mycology. 76:51–167.

Phoulivong S, Cai L, Chen H, McKenzie EHC, Abdelsalam K, Chukeatirote E, Hyde KD. 2010. *Colletotrichum gloeosporioides* is not a common pathogen on tropical fruits. Fungal Diversity. 44:33–43. doi:10.1007/s13225-010-0046-0.

Plan MRR, Joyce DC, Ogle HJ, Johnson GI. 2002. Mango stem-end rot (*Botryosphaeria dothidea*) disease control by partial-pressure infiltration of fungicide. Australian Journal of Experimental Agriculture. 42:625–629. doi:10.1071/EA99123.

Prusky DB, Dann EK, Coates LM. 2022. Postharvest diseases of mango. In: Adaskaveg JE, et al., editors. Postharvest pathology of fruits and nut crops: principles, concepts and management practices. Saint Paul, MN: APS Press; p. 473–486.

Qin LP, Huang SL, Lin SH, Lin CH. 2017. First report of anthracnose of *Mangifera indica* caused by *Colletotrichum siamense* in Sanya City in China. Plant Disease. 101:1038–1038. doi:10.1094/PDIS-09-16-1244-PDN.

Sakalidis ML, Ray JD, Lanoiselet V, Hardy GES, Burgess TI. 2011. Pathogenic Botryosphaeriaceae associated with *Mangifera indica* in the Kimberley Region of Western Australia. European Journal of Plant Pathology. 130:379–391. doi:10.1007/s10658-011-9760-z.

Shivas RG, Tan YP, Edwards J, Dinh Q, Maxwell A, Andjic V, Liberato JR, Anderson C, Beasley DR, Bransgrove K, et al. 2016. *Colletotrichum* species in Australia. Australasian Plant Pathology. 45:447–464. doi:10.1007/s13313-016-0443-2.

Slippers B, Crous PW, Jami F, Groenewald JZ, Wingfield MJ. 2017. Diversity in the Botryosphaeriales: looking back, looking forward. Fungal Biology. 121:307–321. doi:10.1016/j.funbio.2017.02.002.

Slippers B, Johnson GI, Crous PW, Coutinho TA, Wingfield BD, Wingfield MJ. 2005. Phylogenetic and morphological re-evaluation of the *Botryosphaeria* species causing diseases of *Mangifera indica*. Mycologia. 97:99–110. doi:10.1080/15572536.2006.11832843.

Stielow JB, Lévesque CA, Seifert KA, et al. 2015. One fungus, which genes? Development and assessment of universal primers for potential secondary fungal DNA barcodes. Persoonia. 35:242–263. doi:10.3767/003158515X689135.

Tan YP, Shivas RG, Marney TS, et al. 2019. Australian cultures of Botryosphaeriaceae held in Queensland and Victoria plant pathology herbaria revisited. Australasian Plant Pathology. 48:25–34. doi:10.1007/s13313-018-0559-7.

Trakunyingcharoen T, Cheewangkoon R, To-anun C, Crous PW, van Niekerk JM, Lombard L. 2014. Botryosphaeriaceae associated with diseases of mango (*Mangifera indica*). Australasian Plant Pathology. 43:425–438.

Weir BS, Johnston PR, Damm U. 2012. The *Colletotrichum gloeosporioides* species complex. Studies in Mycology. 73:115–180. doi:10.3114/sim0011.

White TJ, Bruns T, Lee S, Taylor J. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis M.A., Gelfand D.H., Sninsky J. J., White T.J., editors. PCR Protocols: a guide to methods and applications. New York: Academic Press; p. 315–322.