

Effects of agricultural practices and fungicides on the management postharvest anthracnose and stem-end rot of mango

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Abstract

Anthracnose and stem-end rot are the main postharvest mango diseases in the Brazilian Northeast. In order to determine the incidence and prevalence of these diseases, near ripe (stage 3) Tommy Atkins fruits were collected from thirty orchards, aged 10 to 12 years old. Inspections and records regarding agricultural practices were undertaken in order to characterize the orchards and evaluate the risk of diseases. Additionally, three experiments were conducted to evaluate different fungicides. Fruits were harvested in Tommy Atkins orchards sprayed with different fungicides and disease incidences were evaluated for two weeks. General averages of incidence and prevalence of stem-end rot were 14.44% and 86.67% respectively, while those of anthracnose were 5.55% and 36.67%. Pearson's chi-squared test identified a significant association between management practices and the occurrence of diseases. The risk of producing diseased fruits is larger in orchards that do not remove residues beneath or above the canopy or diseased panicles and do not adopt good agricultural practices. For orchards that do not adopt good agricultural practices, the relative risk was a 3.82 times higher chance of producing diseased fruits compared to those that adopt good agricultural practices. The fungicides that exhibited efficiency in disease control were pyraclostrobin (0.10 g/L), copper oxychloride (1.60 g/L) and tetraconazole (0.10 g/L). According to the results, in order to control anthracnose and stem-end rot in mangos, producers should spray the orchards with pyraclostrobin, copper oxychloride and tetraconazole fungicides, remove malformed panicles and diseased residues above and beneath the canopy.

Keywords: Botryosphaeriaceae; *Colletotrichum* spp.; Disease control; *Mangifera indica*; Risk analysis.

Introduction

Mango cultivation is one of the most important agricultural activities in Brazilian fruit farming. Mango production in the semi-arid region of the São Francisco Valley meets the internal and international mango market's demand (IBGE, 2022). Climatic factors, water availability and irrigation infrastructure, production technologies and organization of the commercialization chain allowed for the adaptation and success of mango cultivation in this region of Brazil.

Despite the favorable factors for production, mango cultivation is affected by diseases that reduce the productivity and quality of the mango. Stem-end rot and anthracnose are the main diseases that cause economic losses (Terao et al., 2016; Zacaria, 2021), especially during postharvest, which makes disease inspections and control measures difficult in field conditions.

The low adoption rate of cultural control for managing anthracnose and stem-end rot and, specifically, the dearth of fungicides registered by the Ministry of Agriculture (MAPA) for chemical control of stem-end rot are factors that contribute for the incidence of diseases during postharvest (Rehman et al., 2015; Batista et al., 2017; Jenny et al., 2019). The mango debris and the diseased plant material are

important inoculum sources of species of *Colletotrichum* and the fungi *Botryosphaeriaceae* (*Lasiodiplodia theobromae*, *Neofusicoccum parvum* and *Fusicoccum aesculi*), which infect developing fruits (Costa et al., 2010; Lima et al., 2013; Rehman et al., 2015; Batista et al., 2017). These pathogens are commonly found in production orchards associated with dry leaves, mummified fruits, dry branches and panicles (Arauz, 2000; Batista et al., 2017; Noriega-cantu et al., 2017). Periods of high relative humidity or wetness of contaminated crop residues favor the reproduction and accumulation of conidia of the pathogens in the environment (Silva et al., 2009; Batista et al., 2011; Noriega-cantu et al., 2017). Apple crop residues were the main inoculum sources of apple white rot, caused by *F. aesculi* (Valdebenito-Sanhueza et al., 2005).

Measures that avoid infections, such as the reduction of contact of the pathogen with the host plant, creating unfavorable conditions or eradicating the pathogen, are relevant cultural control strategies to reduce the influence of the inoculum source and must be explored to improve the management of diseases. Additionally, to efficiently control diseases in perennials, where the rotation of crops is

impractical, it is necessary to adopt integrated management strategies that involve knowledge of the susceptibility of mango cultivars, favorable environmental conditions, and cultural and chemical control (Coelho et al., 2018; Jenny et al., 2019; Alam et al., 2020; Karunanayake and Adikaram, 2020; Aquino et al., 2021).

The study's objective was to evaluate the incidence of postharvest diseases in commercial areas of the Tommy Atkins cultivar, associate risk factors and identify agricultural practices and fungicides for managing mango postharvest diseases.

Results and Discussion

Survey and effect of agricultural practices on postharvest mango diseases

The diseases associated with postharvest losses were stem-end rot, caused by fungi from the *Botryosphaeriaceae* family, and anthracnose, caused by *Colletotrichum* species. The intensity of the diseases varied greatly between orchards, and places with zero to high incidence of postharvest mango diseases were observed (Figure 1). The general averages of stem-end rot and anthracnose incidences were 14.4% and 5.55%, respectively, while the prevalence of these diseases in orchards was 86.67% and 36.67%, respectively (Figure 1B).

According to the results obtained by Pearson's chi-squared test (Table 1), a significant association between the management practices adopted and the occurrence or non-occurrence of postharvest diseases was observed. Generally, the Relative Risk (RR) of obtaining diseased fruits postharvest is greater in orchards that do not adopt at least one of the studied management practices. Thus, keeping crop residues beneath the plant under the influence of irrigation increases the risk by 1.44 times (CI 95% 1.19-2.03) compared to when residues are removed, while leaving crop residues above the plant's canopy increases the risk by 1.35 times when compared to situations in which said material is removed. A greater risk of disease was also observed when the malformed panicles were kept on the plant's canopy until harvest (RR = 1.55; CI 95% 1.19-2.03) and when the panicle was not cleaned (RR = 2.67; CI 95% 2.06-3.45).

Corroborating the results of the individualized management practices, in orchards that did not adopt good agricultural practices recommended by Integrated Production (IP), which include, among several cultivation practices, monitoring diseases and climate to make decisions regarding disease and pest control; the relative risk of developing postharvest diseases was 3.82 times (CI 95% 2.52-5.80) higher than in orchards that adopt IP's good agricultural practices. Similarly to the RR, the odds of mangos contracting diseases, indicated by the odds ratio (OR) values, were higher in the orchards that did not adopt good agricultural practices.

When evaluating the association between each management practice and the occurrence or non-occurrence of diseases separately (stem-end rot or anthracnose), the chi-squared test was not significant for stem-end rot relative to the practices of removing malformed panicles ($p=0.8361$) and removing crop residues above the plant's canopy ($p=0.4706$). There were also no significant differences for the RR and OR values of both practices, which included value 1 in CI 95% for the RR (0.68-1.36; 0.63-1.23) and OR (0.64,-1.44; 0.58-1.28) (Table 2). For anthracnose, the chi-squared test was not significant for the practice of removing crop residues beneath the plant's canopy ($p=0.0503$) (Table 3).

The results demonstrated that the incidence and prevalence of postharvest diseases in the commercial areas were very different, with some orchards exhibiting good health while others were critical regarding postharvest problems. The incidence and prevalence of plant diseases are epidemiological parameters that vary from season to season, depending on the environmental conditions, the presence of the pathogen and the varieties grown.

Postharvest mango diseases represent a problem for the commercialization of the fruit to the national and international markets. Similarly to what was found in studies in Pakistan (Syed et al., 2017; Alam et al., 2020), stem-end rot's incidence and prevalence were greater compared to anthracnose. This disease has the sensory characteristics of rendering the fruit unfit for consumption and releasing a strong, unpleasant odor in the storage environment. Studying the disease cycle, Alam et al. (2020) determined that *Lasiodiplodia* and *Colletotrichum* were constantly isolated from asymptomatic floral buds, young panicles, senescence flowers and young fruits.

The analyses with risk factors for postharvest mango diseases demonstrated that the differences between the orchards are generally directly associated with phytosanitary management regarding the adoption or non-adoption of agricultural practices that reduce the inoculum source. Inoculum pressure is one of the main causes of epidemic disease control failures (Fiaz et al., 2016; Syed et al., 2017; Alam et al., 2020). In this study, crop residues and diseased material kept in the orchard, underneath or above the plant's canopy, were primary inoculum sources that directly influenced the incidence of postharvest mango diseases.

In addition to the isolated effect of each practice, orchards that adopt IP's good agricultural practices were very important for healthy mango production. The four orchards that did not register any incidence of diseases adopted good agricultural practices and the maximum incidence of postharvest mango diseases recorded on orchards that adopted IP's good agricultural practices was 16.7%. Fiaz et al. (2016), when evaluating the relationship between geographic location and the development of postharvest mango diseases, did not precisely inform the influence of environmental factors and production practices that influenced fruit health. However, they reported the importance of determining factors individually to develop models that make it possible to classify orchards for export means with a low risk of incidence of diseases in the postharvest supply chain. In this work, adopting IP's good agricultural practices was an important reference point for reducing the risks of postharvest diseases.

Generally, the risks of losses to postharvest diseases were associated with the absence of at least one cultural control practice. Only the practices of removing malformed panicles and crop residues above the plant's canopy were not associated with stem-end rot control gains. However, not removing crop residues beneath the plant's canopy increased the risk of this disease. Contradictorily, this was not associated with an increase in the risk of anthracnose.

Botryosphaeriaceae fungi in mango trees, like *Lasiodiplodia*, *Neofusicoccum* and *Fusicoccum* species, are saprophytic and decaying crop residue competitors. Possibly, the moisture created by irrigation over the crop's residues may have favored the proliferation of these pathogens. Studies demonstrated that the reproduction of mango tree pathogens was significantly favored in micro-sprinkler irrigation systems (Silva et al., 2009; Batista et al., 2011). Due to their lifecycle, *Botryosphaeriaceae* depends less on

Table 1. Chi-squared (χ^2), Relative risk (R.R.), Odds ratios (O.R.) and respective confidence intervals at 95% (CI95%) for postharvest diseases (stem-end rot and anthracnose) related to non-adoption of phytosanitary practices in mango production fields of the São Francisco Valley in 2013.

Disease	Variable	Phytosanitary Aspect		χ^2	<i>p</i> -value	R.R. (CI95%)	O.R. (CI95%)
		^a Diseased (%)	^b Healthy (%)				
Anthracnose/ Stem-end rot	Integrated production						
	Does not practice	152(16.89%)	418(46.44%)	51.767	<0.0001*	3.82(2.52-5.80)	4.85(3.05-7.70)
	Practices	23(2.56%)	307(34.11%)				
	Residue beneath canopy						
	Does not remove	334(37.11%)	391(43.44%)	11.363	0.0007*	1.44(1.14-1.81)	1.81(1.28-2.57)
	Removes	56(6.22%)	119(13.22%)				
	Malformed panicle						
	Does not remove	70(7.78%)	200(22.22%)	10.344	0.0013*	1.55(1.19-2.03)	1.75(1.24-2.46)
	Removes	105(11.67%)	525(58.33%)				
	Residue above canopy						
	Does not remove	77(8.56%)	253(28.11%)	5.030	0.0249*	1.35(1.04-1.77)	1.46(1.04-2.05)
	Removes	98(10.89%)	472(52.44%)				
	Cleans panicle						
	Does not clean	61(6.78%)	89(9.89%)	51.756	<0.0001*	2.67(2.06-3.45)	3.82(2.61-5.60)
Cleans	114(12.67%)	636(70.67%)					

^aDiseased (%): number of diseased fruits and (percentage); ^bHealthy (%): number of healthy fruits and (percentage). *Significant at *p*-value <0.05.

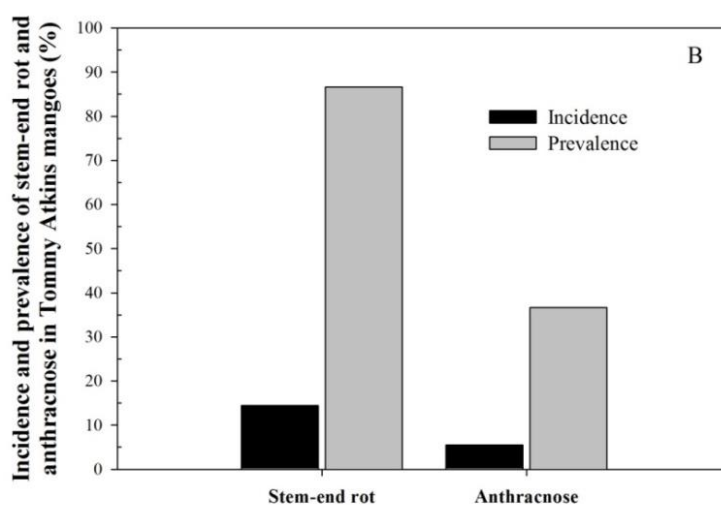
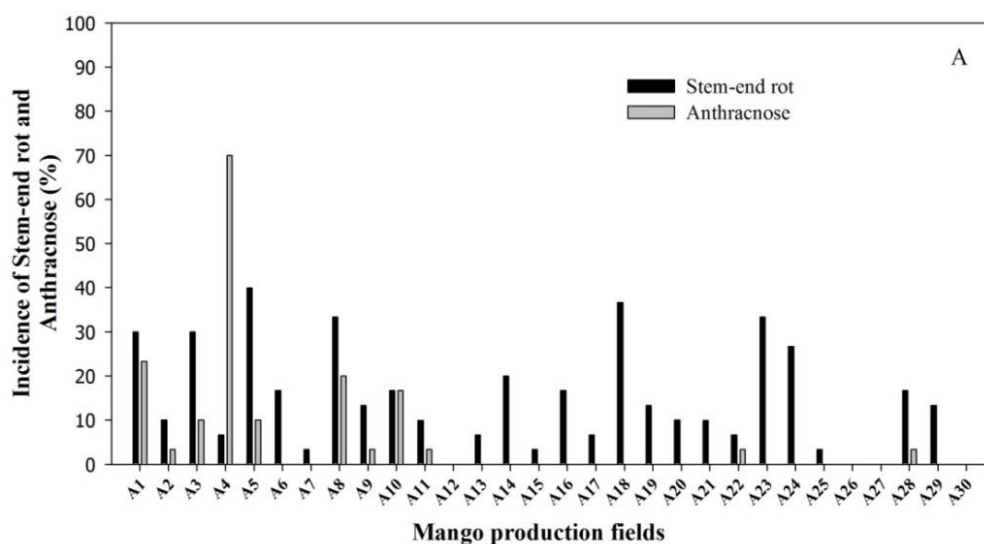


Fig 1. Incidence of stem-end rot and anthracnose diseases in different 'Tommy Atkins' mango production fields (A1 to A30) in the São Francisco Valley in 2013 (1A); and overall averages of incidences and prevalence of these postharvest diseases (1B).

Table 2. Chi-squared (χ^2), Relative risk (R.R.), Odds ratios (O.R.) and respective confidence intervals at 95% (CI95%) for mango stem-end rot related to non-adoption of phytosanitary practices in mango production fields of the São Francisco Valley in 2013.

Disease	Variable	Phytosanitary Aspect		χ^2	<i>p</i> -value	R.R. (CI95%)	O.R. (CI95%)
		^a Diseased (%)	^b Healthy (%)				
Stem-end rot	Integrated production						
	Does not practice	108(12%)	462(51.33%)	25.506	<0.0001*	2.84(1.83-4.40)	3.27(2.02-5.29)
	Practices	22(2.44%)	308(34.22%)				
	Residue beneath canopy						
	Does not remove	84(9.33%)	426(47.33%)	3.909	0.048*	1.39(1.0-1.95)	1.47(1.0-2.17)
	Removes	46(5.11%)	344(38.22%)				
	Malformed panicle						
	Does not remove	38(4.22%)	232(25.78%)	0.042	0.8361 ^{NS}	0.96(0.68-1.36)	0.96(0.64-1.44)
	Removes	92(10.22%)	538(59.78%)				
	Residue above canopy						
Does not remove	44(4.89%)	286(31.78%)	0.520	0.4706 ^{NS}	0.88(0.63-1.23)	0.86(0.58-1.28)	
Removes	98(9.56%)	484(53.78%)					
Cleans panicle							
Does not clean	33(3.67%)	117(13.00%)	8.314	0.0039*	1.70(1.19-2.42)	1.90(1.22-2.95)	
Cleans	97(10.78%)	653(72.56%)					

^aDiseased (%): number of diseased fruits and (percentage); ^bHealthy (%): number of healthy fruits and (percentage). *Significant at *p*-value <0.05. ^{NS}not significant at *p*-value <0.05.

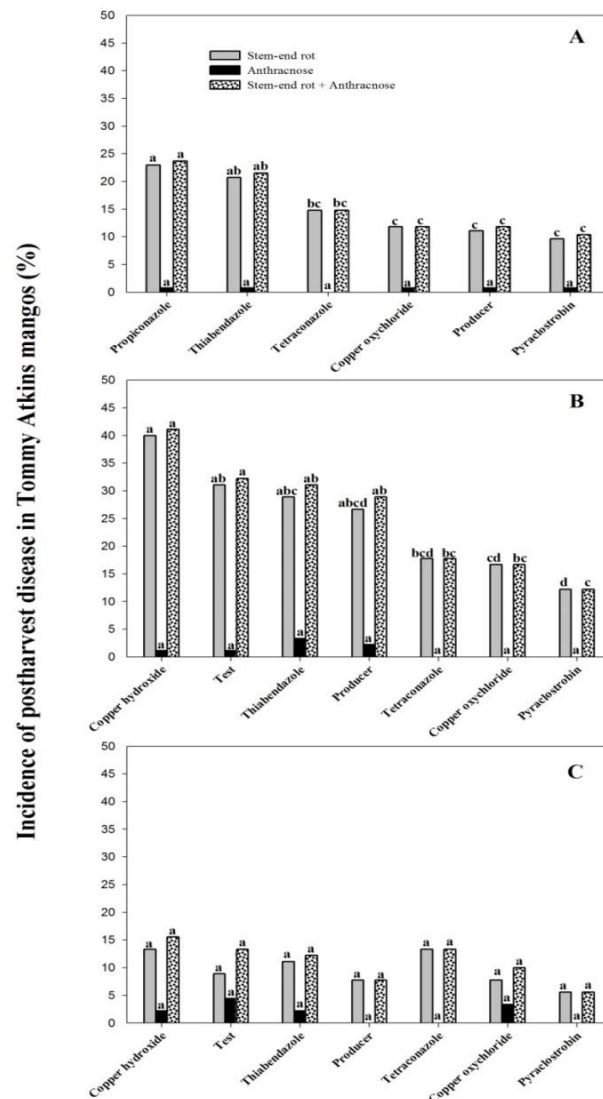


Fig 2. Effect of preharvest fungicide treatments on total incidence of postharvest disease in Tommy Atkins mangos (stem-end rot and anthracnose) in three commercial production fields in the 2014 (A) – 2016 (B and C) period. Means followed by the same letter do not differ according to Duncan's test at $\alpha=0.05$.

Table 3. Chi-squared (χ^2), Relative risk (R.R.), Odds ratios (O.R.) and respective confidence intervals at 95% (CI95%) for mango anthracnose relating to non-adoption of phytosanitary practices in mango production fields of São Francisco Valley in 2013.

Disease	Variable	Phytosanitary Aspect		χ^2	p-value	R.R. (CI95%)	O.R. (CI95%)
		^a Diseased (%)	^b Healthy (%)				
Anthracnose	Integrated production						
	Does not practice	48(5.33%)	522(58.0%)	24.327	<0.0001*	13.89(3.39-56.79)	15.08(3.64-62.46)
	Practices	2(0.22%)	328(36.44%)				
	Residue beneath canopy						
	Does not remove	35(3.89%)	475(52.78%)	3.832	0.0503 ^{NS}	1.78(0.98-3.22)	1.84(0.99-3.42)
	Removes	15(1.67%)	375(41.67%)				
	Malformed panicle						
	Does not remove	35(3.89%)	235(26.11%)	40.336	<0.0001*	5.44(3.02-9.80)	6.10(3.27-11.38)
	Removes	15(1.67%)	615(68.33%)				
	Residue above canopy						
	Does not remove	36(4.0%)	294(32.67%)	28.461	<0.0001*	4.44(2.43-8.11)	4.86(2.58-9.16)
	Removes	14(1.56%)	556(61.78%)				
	Cleans panicle						
	Does not clean	28(3.11%)	122(13.56%)	58.972	<0.0001*	6.36(3.74-10.81)	7.59(4.21-13.70)
Cleans	22(2.44%)	728(80.89%)					

^aDiseased (%): number of diseased fruits and (percentage); ^bHealthy (%): number of healthy fruits and (percentage). *Significant at p-value <0.05. ^{NS}not significant at p-value <0.05.

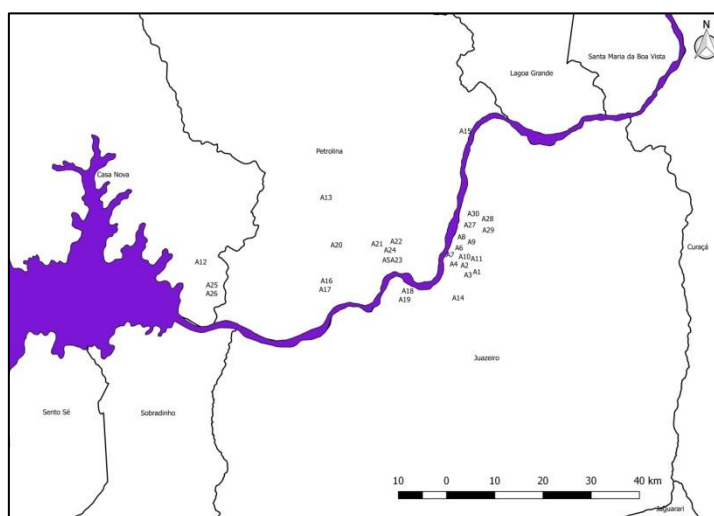


Fig 3. Location of the 'Tommy Atkins' mango production fields (A1 to A30) sampled in the São Francisco Valley (Petrolina, Pernambuco State; Juazeiro and Casa Nova, Bahia State) in 2013.

external factors (temperature and humidity) to cause postharvest stem-end rot. According to Johnson et al. (1992), endophytic colonization of the inflorescence and pedicel is the primary route of infection, as the fungi associated with stem-end rot occur endophytically in mango tree branches well before the emergence of the inflorescence. This phenomenon partly explains why leaving crop residues above the plant's canopy has little influence on the occurrence of disease, unlike not cleaning the panicles. In the latter situation, young fruits commonly affected by stem-end rot in their juvenile state contaminate nearby fruits (Batista et al., 2017) and must be removed from the panicles. On the other hand, the environment created by the malformed panicle favored the occurrence of anthracnose. The species of *Colletotrichum* are well known

for the ability to cause a complete burn of the mango tree's inflorescence, infecting, in addition to the floral set, the peduncle, rachis and pedicel (Terao et al., 2016; Tovar-Pedraza et al., 2020).

Efficiency of fungicide sprays in field conditions for controlling postharvest mango diseases

According to the chemical control studies, differences were observed among fungicides in controlling postharvest mango diseases (Figure 2). Only in experiment three did the fungicides not exhibit significant differences between them, probably related to a lower disease incidence (Figure 2C). There was no significant difference between treatments from three experiments evaluated for anthracnose and the incidence of the disease in the experiments was of less than

five percent (5%). However, comparing the treatments in experiments one and two, there were significant differences between treatments of fungicides to control stem-end rot and stem-end rot+anthracnose. The first experiment indicated that treatments with pyraclostrobin, producer and copper oxychloride were significantly different from propiconazole and thiabendazole, but not from tetraconazole (Figure 2A). The treatments with pyraclostrobin, copper oxychloride and tetraconazole provided excellent control against postharvest diseases (stem-end rot+anthracnose) compared to the Test treatment in experiment two (Figure 2B). Surprisingly, the effect of the tetraconazole fungicide was not significant when stem-end rot disease was analyzed separately. The pyraclostrobin fungicide was the most efficient in postharvest disease control.

Strobirulins have been used with success in the postharvest control of mango fruit diseases (Manasa et al., 2018; Alam et al., 2020; Rehman et al., 2020). Studies with different fungicides for controlling mango diseases are important, since some fungicides are being banned in several countries, like prochloraz in the European Union. Based on these results, control of these diseases may be achieved with alternating applications of the fungicides tetraconazole, copper oxychloride and pyraclostrobin. While tetraconazole acts by inhibiting the biosynthesis of ergosterol, pyraclostrobin interferes in mitochondrial respiration and copper oxychloride possesses a nonspecific action mechanism (Ghini and Kimati, 2002). This strategy has the advantage of reducing the risks of selection of resistant isolates among the pathogens involved with both diseases, as the three fungicides have different sites of action on the pathogens.

Materials and Methods

Survey and effect of agricultural practices on postharvest mango diseases

In March 2013, Tommy Atkins mango cultivar fruits were collected from different commercial orchards in the cities of Petrolina-PE, Casa Nova-BA and Juazeiro-BA, in the Northeast of Brazil (Figure 3). Thirty orchards (10 to 12 years old) were evaluated, of which 30 mangos were collected that did not exhibit physical or biological damage and were in ripening stage 3 (Assis and Lima, 2008), generating 900 fruits. The orchards were sampled for two weeks and georeferenced with GPS (global positioning system) instruments. The samples were identified and submitted to the laboratory, where the fruits were washed with water and neutral soap, rinsed in running water, air-dried and kept in cardboard boxes at a room temperature of 25°C for two weeks. At the end of this period, the incidence of anthracnose and stem-end rot symptoms was evaluated. Disease incidence was surveyed at 15 days of storage and calculated as follows: Disease incidence = (the number of diseased mangos/total number of mangos) x 100%. The prevalence of each disease was calculated as the total number of mango production fields with diseases (anthracnose or stem-end rot) divided by the total number of mango production fields (sampling fields) and expressed as a percentage.

The orchards visited are part of the irrigation projects complex of Petrolina-PE (Nilo Coelho and Bebedouro), Juazeiro-BA (Mandacaru, Maniçoba and Tourão) and Casa Nova-BA. In addition to the mango collections, records were made, in forms, of the presence or absence of agricultural

control practices: a) crop residues above the plant's canopy; b) crop residue beneath the plant's canopy; c) malformed panicles; d) cleaned panicles; e) type of commercial production (Conventional or Integrated Production - IP). Integrated Production - IP corresponds to those mango production fields that adopt the good agricultural practices foreseen in Brazilian normative ruling (Instrução Normativa - Sarc) nº 012, from September 18th, 2003.

Efficiency of fungicide sprays in field conditions for controlling postharvest mango diseases

Three experiments were conducted in the period between 2014 and 2016 in orchards from the Tommy Atkins cultivar (12 years old) with a history of anthracnose and stem-end rot postharvest mango diseases and selected among the orchards from the previous survey. The planting spacing was of 8x5 meters in the three production fields.

The treatments tested in the first experiment (2014) were: 1) copper oxychloride (1.60 g/L); 2) pyraclostrobin (0.10 g/L); 3) thiabendazole (0.485 g/L); 4) tetraconazole (0.10 g/L); 5) propiconazole (0.125 g/L); and 6) control carried out by the producer. In the second and third experiments, the treatments tested were: 1) copper oxychloride (1.60 g/L); 2) copper hydroxide (1.076 g/L); 3) pyraclostrobin (0.10 g/L); 4) thiabendazole (0.485 g/L) and 5) tetraconazole (0.10 g/L); 6) control carried out by the producer; 7) Test without fungicide (0.0 g/L). The producer's treatments were two applications of thiabendazole (0.485 g/L), two of difenoconazole (0.125 g/L) and one of copper hydroxide (1.076 g/L). The sprayings were carried out using a turbo atomizer attached to a tractor and the quality of spray coverage was measured using water-sensitive paper in order to obtain a droplet density above 70/cm². The spray water's pH was adjusted to pH 6.0.

Five sprayings were carried out, the first during the ripening of branches and the remainder as flowering began, biweekly. During the harvest, 45 fruits (experiment 1) and 30 fruits (experiments 2 and 3) were randomly collected per repetition. The fruits were cleaned and packaged as in the activity in the previous experiment, and evaluated after two weeks for the incidence of symptoms of anthracnose and stem-end rot. The experiments were set up in a randomized block design, with three repetitions and 26 plants per plot in each treatment.

Statistical analysis

The collected data from the different production fields (A1 to A30) were used to compare the proportions of healthy and diseased fruits relative to management practices. To identify associations between different management practices and the occurrence of diseases, Pearson's chi-squared test and the measures of association of relative risk (RR) and odds ratio (OR) and their respective intervals with 95% confidence were used. For each fungicide experiment, the values for final disease incidence were analyzed using ANOVA and, when necessary, means were compared using Duncan's multiple range test (P = 0.05). The SAS System was used for all analyses.

Conclusions

According to the results, associating cultural management measures to reduce inoculum sources with fungicides results in the reduction of the postharvest diseases stem-end rot and anthracnose. Therefore, the adoption of good

agricultural practices, including different cultural control measures, along with chemical control with fungicides constitutes the best strategy, in this study, for managing these diseases.

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