Scientific Research and Essays Vol. 5(17), pp. 2400-2403, 4 September, 2010
Available online at http://www.academicjournals.org/SRE
ISSN 1992-2248 ©2010 Academic Journals

Full Length Research Paper

Studies on microorganisms associated with pre-harvest deterioration of guava (*Psidium guajava* Linn.) fruits

A. A. Ajayi[1]*, S. C. Yah[2], G. I. Olasehinde[1] and O. O. Ayepola[1]

[1]Department of Biological Sciences, College of Science and Technology, Covenant University, Ota, Ogun State, Nigeria.
[2]University of Witwatersrand, Johannesburg, South Africa.

Accepted 4 June, 2010

Women form the largest percentage of sellers and buyers in markets all over the world. There are lots of benefits to be derived from guava (*Psidium guajava* Linn.) fruits but a large percentage of guava fruits produced annually are lost to pre harvest deterioration caused by microorganisms. Four fungal pathogens, *Aspergillus niger, Rhizopus* sp*., Fusarium* sp*., Penicillium* sp. and yeast cells were found to be associated with pre-harvest deterioration of Guava (*P. guajava* Linn.) in Ota, Ogun State, Southwestern Nigeria. *A. niger, Penicillium* sp. and yeast cells were the most prevalent while *Penicillium* sp. was the most pathogenic, inducing a rot of 61 mm in diameter within seven days of incubation. The ash and moisture contents of the uninfected fruits were higher than that of the infected ones. The results of this present investigation could be utilized in juice making industries in Nigeria as well as for the local women who deal directly with the selling and buying of these fruits in our markets.

Key words: *Psidium guajava* Linn., fungal pathogens, pre-harvest deterioration.

INTRODUCTION

Guava (*Psidium guajava* Linn) belongs to the family Myrtaceae and it thrives both in humid and dry climates (Morton, 1987). A lot of medicinal values can be derived from the leaves, bark and fruit of guava (Abdelrahim et al., 2002). In some parts of the world, the plant is believed to be very useful in the treatment of diarrhea, gastroenteritis, intestinal worms, gastric disorders, vomiting, coughs, vaginal discharges, menstrual pain, haemorrhages and edema (Jairaj et al., 1999; Lozoya et al., 2002; Hsieh et al., 2005; Lutterodt and Maleque, 1988). Guava is enriched in tannins, phenols, triterpenes, flavonoids, essential oils, saponins, carotenoids, lectins, vitamins, fiber and fatty acids (Padula and Rodriguez, 1986). Guava leaf extracts and fruit juice is very good in the cure of infantile rotaviral entities (Wei et al., 2000). Guava fruit contains high amount of vitamin A and it is higher in vitamin C than citrus as it contains about 80 mg of vitamin C in 100 g of fruit (Suntornsut et al., 2002). Despite all the benefits that can be derived from guava fruits, leaves, barks and other parts of a guava tree, a very high percentage is lost to pre-harvest deterioration caused by microorganisms. Guava fruits obtained from private farms as well as on the Covenant University Campus, Ota, Ogun State, revealed the presence of several fruits deteriorating on the trees. This study therefore, investigated the etiology of pre-harvest deterioration of guava fruits and its effects on the quality of the fruits in Ota, Ogun State, South Western, Nigeria.

*Corresponding author. E-mail: quietasever@yahoo.com.

MATERIALS AND METHODS

Collection of samples

Infected and non-infected guava fruits obtained from private farms and trees on the Covenant University Campus were kept in sterile sample bags and taken to the laboratory of the Department of Biological Sciences, Covenant University, Ota, Ogun State, Nigeria for analysis.

Methods

Cultural conditions

Fruits were carefully separated, infected fruits from non-infected fruits. The infected portions were excised and cut into 2 × 2 mm pieces, surface sterilized with 1% Sodium Hypoclorite solution (NaOCl) and rinsed in four successive changes of sterile distilled water to remove the residual effect of the Sodium hypoclorite

Table 1. Incidence of Occurrence of Fungi found associated with guava (*P. guajava* L.) deterioration.

Fungal isolates	Rate of occurrence (%)	Diameter of infected area in millimeter (X ± S.D)
A. niger	100	5.8 ± 1.4
Fusarium species	95	6.7 ± 2.8
Rhizopus species	82	36.4 ± 3.4
Penicillium species	100	61 ± 12.0
Yeast cells	100	16.0 ± 11.3

Table 2. Moisture and ash content of infected and non-infected guava (*P. guajava* L.) fruits.

Sources	Type	Moisture (g)	Ash (g)
CU campus, Ota	Non infected	36.6	1.21
CU campus, Ota	Infected	34.5	1.11
Private farms, Ota	Non infected	25.9	1.10
Private farms, Ota	Infected	24.7	1.01

solution. The excised infected portions were then plated on to Sabouraud dextrose agar in petri dishes and incubated for six days under alternating 12-h light and dark periods at 26°C. Fungal cultures were examined under the microscope. The identity of these fungi was certified using cultural, morphological, pathogenicity tests as well as comparing them with confirmed representatives of the different species (Barnett and Hunter, 1972; Booth, 1977; Purseglove, 1968).

Moisture and ash content test

The infected and non-infected fruits per tree were weighed and lined in a hot oven at 60°C for ten (10) days. The dried pieces of the fruits were ground into powder and analyzed for Moisture and Ash content according to the AOAC (1984) procedure.

Establishment of pathogenicity

Freshly harvested fruits were surface sterilized as described above. A sterile cork borer (4 mm) was used to remove a tissue disc from each of the fruits prior to inoculation with the fungal isolates. Discs (4 mm) each obtained from the edge of a 96-h-old culture of the fungal isolates were used to inoculate each of the fruits. The points of inoculation were sealed with the scooped out tissues and it was sealed with molten wax. Control fruits were similarly treated except that sterile Sabouraud dextrose agar disc served as the inoculum. The inoculated fruits were then kept in polythene bags containing moist cotton wool to maintain high relative humidity and it was incubated at room temperature of 25°C for 7 days. The extent of rot was determined by measuring the size of infection. Wet mounts of the hyphal structures obtained from the infected fruits were stained with lactophenol in cotton blue and viewed under the compound microscope for the presence of the pathogens. They were identified as described above.

RESULTS

Four fungal isolates and yeast cells were found to be associated with pre-harvest deterioration of guava fruits. The fungi were *Aspergillus niger, Rhizopus* sp., *Fusarium* sp. and *Penicillium* sp. *Penicillium* sp. was the most pathogenic, inducing a rot of 61 mm in diameter within seven days of incubation. *A. niger, Penicillium* sp. and yeast cells were the most prevalent (Table 1). The Ash and moisture content was higher in the non-infected fruits than the infected ones (Table 2).

DISCUSSION

Previous researchers have reported the involvement of fungi in pre harvest as well as post harvest deterioration of many tropical fruits (Adisa and Fajola, 1982; Olunloyo, 1979; Adejuwon and Olutiola, 2005). *A. niger* has been reported in post harvest deterioration of tomatoes fruits (Ajayi and Olasehinde, 2009). *Aspergillus flavus* Linn, *Rhizopus arrhizus* Fisher and *Botryodiplodia theobromae* Pat. were implicated in post harvest deterioration of tomato fruits by fungi (Ajayi et al., 2003; Ajayi et al., 2007a and b). *Penicillium citrinium* and *Penicillium steckii* has previously been reported in the deterioration of cocoa beans (Olutiola, 1982, 1983). The Pre-harvest rot of Sour sop was associated with *A. niger* and *Rhizopus stolonifer* (Amusa et al., 2003). The Pathogenicity test revealed that all the isolates were pathogenic (Tables 3 - 7). *Penicillium* sp. induces a rot of 61 mm in diameter within seven days of incubation while *Rhizopus* sp. induces a rot of 36 mm in diameter within the same period of time (Table 1). The Ash and moisture contents of the infected fruits were a little lower than that of the non-infected ones (Table 2). It could be deduced that this reduction in moisture content will definitely affect the protein and the carbohydrate content of the fruit apart from the unfavorable physical conditions of the fruit caused by the microorganisms. The

Table 3. Pathogenicity test of guava (*P. guajava* L.) fruits due to infection by *A. niger.*

Types of guava	D1	D2	D3	D4	D5	D6	D7
G1	4.0	4.2	4.5	5.1	5.8	6.3	6.7
G2	4.0	4.4	4.5	4.8	5.6	6.4	6.8
G3	4.0	4.1	4.6	5.3	6.1	6.7	7.4
G4	4.0	4.4	4.7	5.6	5.9	6.3	7.0
G5	4.0	4.3	4.5	5.8	6.0	6.3	7.1
G6	4.0	4.3	4.5	4.9	5.8	6.4	7.5
G7	4.0	4.3	4.6	4.9	5.9	6.2	7.9
G8	4.0	4.2	4.4	5.0	6.3	6.7	7.6
Gc9	4.0	4.0	4.0	4.0	4.0	4.0	4.1

Key: D = days, G = guava infected, GC = Control (inoculated with only PDA alone).

Table 4. Pathogenicity test of Guava (*P. guajava* L.) fruits measured in millimeters (4 mm) due to infection by *Fusarium* species.

Types of guava	D1	D2	D3	D4	D5	D6	D7
G1	4.0	4.4	4.6	5.3	5.9	6.4	6.9
G2	4.0	4.3	4.7	5.8	6.6	7.3	7.8
G3	4.0	4.5	5.0	5.0	6.6	7.4	7.7
G4	4.0	4.3	4.8	5.5	5.9	6.7	7.2
G5	4.0	4.1	4.3	4.8	5.0	6.4	6.7
G6	4.0	4.1	4.6	5.0	5.4	5.8	6.3
G7	4.0	4.2	4.3	4.8	5.3	5.8	6.1
G8	4.0	4.0	4.1	4.5	4.9	5.3	5.5
Gc9	4.0	4.0	4.0	4.0	4.0	4.1	4.2

Key: D = days, G = guava infected, GC = Control (inoculated with only PDA).

Table 5. Pathogenicity test of Guava (*P. guajava* L.) fruits measured in millimeters (4 mm) due to infection by *Rhizopus* species.

Types of guava	D1	D2	D3	D4	D%	D6	D7
G1	4.0	4.0	4.4	5.3	6.1	6.3	6.5
G2	4.0	4.3	4.7	4.8	5.3	6.2	6.5
G3	4.0	4.3	4.6	4.7	6.4	6.9	7.3
G4	4.0	4.1	4.5	5.1	5.6	6.7	7.1
G5	4.0	4.1	4.7	5.9	6.6	7.3	7.8
G6	4.0	4.3	4.7	5.3	5.7	6.9	7.2
G7	4.0	4.5	4.8	5.4	5.7	6.5	7.4
G8	4.0	4.2	4.3	4.6	5.7	5.9	6.3
Gc9	4.0	4.0	4.0	4.0	4.0	4.0	4.0

Key: D = days, G = guava infected, GC = Control (inoculated with only PDA).

Table 6. Pathogenicity test of Guava (*P. guajava* L.) fruits measured in millimeters (4 mm) due to infection by *Penicillium* species.

Types of guava	D1	D2	D3	D4	D5	D6	D7
G1	4.0	4.3	4.5	4.8	5.3	5.9	6.3
G2	4.0	4.1	4.9	5.6	6.0	6.7	7.7
G3	4.0	4.1	4.2	4.7	5.3	5.8	6.5
G4	4.0	4.2	4.4	4.7	5.5	6.2	7.2

Table 6. Contd.

Types of guava	D1	D2	D3	D4	D5	D6	D7
G5	4.0	4.1	4.3	5.1	5.7	6.1	6.7
G6	4.0	4.3	4.7	5.3	5.7	6.9	7.2
G7	4.0	4.5	4.8	5.4	5.7	6.5	7.4
G8	4.0	4.1	4.2	4.9	5.6	6.0	6.7
Gc9	4.0	4.0	4.0	4.0	4.0	4.0	4.0

Keys: D = days, G= guava infected, GC = control (inoculated with only PDA).

Table 7. Pathogenicity test of guava (*P. guajava* L.) fruits measured in millimeters (Coker borer (4 mm) due to yeast cells.

Types of guava	D1	D2	D3	D4	D5	D6	D7
G1	4.0	4.1	4.3	4.5	4.9	5.4	6.0
G2	4.0	4.1	4.4	4.7	5.3	5.7	6.1
G3	4.0	4.1	4.3	4.7	5.1	5.8	6.6
G4	4.0	4.1	4.2	4.3	4.8	5.1	5.2
G5	4.0	4.1	4.2	4.4	4.5	4.9	5.1
G6	4.0	4.0	4.1	4.3	4.6	4.9	5.0
G7	4.0	4.1	4.2	4.4	4.7	4.8	4.9
G8	4.0	4.0	4.2	4.5	4.8	5.0	5.3
Gc9	4.0	4.0	4.0	4.0	4.0	4.0	4.0

Key: D= days, G= guava infected, GC= control (inoculated with only PDA).

decrease in moisture and ash content could be due to degradative activities of the pathogens associated with the fruit. This will also reduce the quality of the fruit and reduce the percentage of annual production of guava despite all its benefits if not addressed.

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