
Decay of Guava Fruit (*Psidium guajava* Linn.) Quality Caused by Some Mold Fungi.

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Abstract Guava (*Psidium guajava* Linn.) is a very popular fruit; it is generally a good source of lycopene, beta-carotene, vitamin C, protein, fat, carbohydrate, fibers, minerals, vitamin B & B₂ and is an excellent source. Also, Guava is one of the most liked fruit items in Egypt and has its own economical importance. The most important causal agent responsible for the post-harvest diseases of Guava, are the fungi. These microorganisms invade the fruit and cause considerable damage at the post-harvest stage, during transit, storage and transportation to the market. The isolation of post harvest pathogen from diseased guava fruits resulted that, one hundred and eighty fungal colonies were isolated from three different Governorates (Localities), in Egypt i. e. Beheira (44.44%), El-Sharkia (38.89%) and Qualubia (16.67%). Four fungal genera belonging to six species were identified. These are *Aspergillus* (*A. flavus* (26.67%), *A. niger* (7.78%) and *A. parasiticus* (3.33%), *Botryodiplodia theobrome* (17.22%), *Fusarium oxysporum* (2.22%) and *Rhizopus stolonifer* which was higher fungal frequency (42.78%). Aflatoxins were detected with *Aspergillus parasiticus* only. Aflatoxin G₁ was detected with isolate No. 8 *A. parasiticus* from Qualubia sample which record 0.548 ng/ml. While isolate No. 10 from Beheira sample gave higher aflatoxins AFB₁ and AFG₁ which recorded 0.163 ng/ml and 0.296 ng/ml respectively. All tested fungi i. e. *A. flavus*, *A. parasiticus*, *B. theobrome*, *F. oxysporum* and *R. stolonifer* were found to be decreased all determined of physical and chemical properties i.e. fresh weight (g), total soluble solids (TSS%), total titratable acidity (TA%), TSS/TA ratio % and Ascorbic acid (mg/100g of fruit weight) compared with un-infected Guava fruits. Increasing reduction as well as percentage of loss and decreased postharvest shelf life on marketable period by all tested fungi with increasing the storage period from one to two weeks.

Keywords: Guava rotted fruits, Fungi, Mycotoxins, Fruit quality

Introduction

Guava (*Psidium guajava* Linn., Family: Myrtaceae) fruit is a berry with a large seedy core. Guava is a large dicotyledonous shrub, or small evergreen tree. The pulp inside may be sweet or sour, and off-white ("white" guavas) to deep pink ("red" guavas). The seeds in the central pulp vary in number and hardness,

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depending on species. Guava is enriched in tannins, phenols, triterpenes, flavonoids, essential oils, saponins, carotenoids, lectins, vitamins, fiber and fatty acids. Guava leaf extracts and fruit juice is very good in the cure of infantile rotaviral entities. 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 (Wei *et al.*, 2000, Suntornsut *et al.*, 2002 and Misra, 2004).

The fruit may be smooth or ridgy and waxy. Presently guava is being grown all over the sub-tropical and tropical world, due to its high dietary value and good flavor. Guava fruit contains high amounts of Vitamins A, B₁ (Thiamin), B₂ (Riboflavin) and C. It is a rich source of vitamin C (Ascorbic acid). The vitamin C contents of Guava fruit are four times higher than those of citrus. Guava is commercially picked when it starts turning from green to yellow so that it ripens one day later in the transit before marketing (Bokhari 2009 and Ajayi *et al.*, 2010). In case of ascorbic acid, pectin and other minerals contents it scores high over other fruit; that is why the common Guava is aptly referred to as “poor man’s apple” and / or “apple of the tropics”. Guava is a very productive and highly profitable fruit crop. Guava can grow in many types of soil and it can grow under a wide range of climatic and soil conditions and can tolerate alkaline soil up to pH 8.2 (Mathew, 2010). In Egypt, Guava trees are widely planted especially in Beheira, El-Sharkia, around Alexandria and newly reclaimed lands. Guavas occupy about 38000 feddan, yielded about 314000 ton as annual fruit production with an exported range about 16.312.38 metric tons to many countries. Guava exports from Egypt are increased through air flight as the main transport system (Omayma M. Ismail *et al.*, 2010).

The principle of spread of fungal infection in fruits supports that a single infected Guava fruit can be the source of infection to other guava fruits during storage and in transit. Guava fruits constitute a vital part of human diet. Microorganisms are associated, in a variety of ways with all the foods we eat, Guava fruits inclusive (Jay, 2003 and Misra, 2004). Fruit rot and postharvest diseases are important and cause serious losses. Around 90-100 percent fruits have been found infected with several fungi namely *Pestalotia psidii*, *Colletotrichum gloeosporioides*, *Rhizopus stolonifer* and *Aspergillus niger* during storage. Fungal infection on the fruit may occur during the growing season, harvesting, handling, transport and post-harvest storage and marketing conditions, or after purchasing by the consumer (Nongmaithem, N. 2014 and Amadi *et. al.*, 2014). A total of seven (7) fungi were isolated from the postharvest spoilage of Guava fruit namely *Colletotrichum gloeosporioides*, *Fusarium oxysporum*, *Mucor sp.*, *Rhizopus stolonifer*, *Aspergillus niger*, *A. fumigatus* and *A. parasiticus*. *Fusarium oxysporum* was the most prevalent of the seven fungi isolated and appeared in all the four locations. *Aspergillus soft*

rot is caused by several species of *Aspergillus* of which *A. awamori*, *A. wentii* and *A. niger* are important (Adisa, 1985 and Misra, 2004). It is estimated that about 20-25% of the harvested fruits are decayed by pathogens during post-harvest handling even in developed countries (Droby, 2006 and Zhu, 2006). 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.).

Aspergillus niger, *Penicillium* sp. and yeast cells were the most prevalent while *Penicillium* sp. was the most pathogenic. The common postharvest and storage fungi of fruits are *Alternaria* spp., *Aspergillus* spp., *Fusarium* spp., and *Penicillium* spp. (Bhale, 2011). Besides the losses in income to the fruit marketers, in some cases host pathogen interactions provide a favorable environment and source for production of many different compounds. Mycotoxins are produced by several genera in plants during the growing season when portals of entry are provided and environmental conditions are appropriate and be continued or initiated in postharvest and stored products. The majority of these toxins are produced by fungi of the genera, *Aspergillus*, *Penicillium* and *Fusarium* (Barkai-Golan, Zain, 2011 and Ammar and El-Naggar, 2014). Thus, the presence of fungi is a serious health hazard for workers as well as consumers in markets. It is crucial for the post-harvest quality management of a wide range of high value fruit crops Pande *et al.* (2012), Sarmah, and Sarma (2012) and Vermani *et al.* (2014). The storage fungi, primarily species of *Aspergillus* and *Penicillium* also grow well at lower moisture contents (Ammar and El-Naggar, 2014). In Egypt under local markets, there is relatively little information related to the natural occurrence of fungi and mycotoxins in fruits.

The Present Study Includes: 1-Survey of some fungal plant diseases, isolation and identification the association of fungal diseases with Guava roted fruits, 2- Tested of mycotoxins production, 3- Study the changes in fruit quality i.e. a- total soluble solids (TSS%), b- total titratable acidity (TA%), c- TSS/TA ratio % and d- Ascorbic acid mg/100g .

Material and methods

Samples Collection

A survey of crop fungi was conducted on the economically important fruits of Guava during 2013/2014 season. Naturally infected of mature yellowish-green Guava fruits were collected from three orchards as well as from local markets at Beheira, El-Sharkia and Qualubia governorates, Egypt.

Samples were brought to the laboratory in separate sterilized polythene bags (Ammar and El-Naggar, 2014).

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 Hypochlorite solution (NaOCl) for 1 min and rinsed in sterile distilled water to remove the residual effect of the Sodium hypochlorite solution, then plated on sterile potato dextrose agar (PDA) in Petri dishes and incubated for six days under alternating 12 hr light and dark periods at 25±2 °C (Ammar, and El-Naggar, (2014)). Fungal hyphae, growing out from the infected fruit pieces were purified on PDA slants. Pure culture was maintained by periodic sub culturing. Fungal cultures were examined under the light microscope in the National Research Centre (NRC), Plant Pathology Dept., Egypt. The identity of these fungi was certified using cultural and morphological characteristics' with the help of available literature i. e. Raper and Fennell (1965); Smith (1969); Booth (1977); Biligrani *et al.*, (1991) and Barnett and Hunter (1999).

Tested of mycotoxin production

The different mycotoxigenic fungal isolates (*Aspergillus flavus* A. *parasiticus* and *Fusarium oxysporum*) were propagated as pure culture in 100 ml (SMKY) broth (Sucrose 200 g, MgSO₄·7H₂O 0.5 g, KNO₃ 3 g, yeast extract 7 g) and incubated in dark condition at 26±2 °C for 15 days in Food Toxicology and Contamination Dept., National Research Centre (NRC). After incubated period, were prepared for toxin determined by HPLC. The determination of aflatoxins was carried out by using HPLC according to (AOAC, 2007). The HPLC instrument used was waters (474) system, equipped with quaternary pump. The fluorescence detector system was set at 360 nm excitation and 440 nm emission wavelengths. The chromatography column was phenomenex c18 (250x 4.6 mm), 5 µm. The mobile phase system (H₂O: MeOH: CH₃CN, 30:60:10 v/v/v) was isocratically at flow rate of 1 ml/min. The data were collected and integrated using Totalchrom Navigator Chromatography Manager Software according AOAC (2007); Han *et al.* (2004) and Embaby *et al.* (2007 and 2012).

Fruit Quality

Healthy apparent of fresh (mature yellowish-green) Guava fruits were contaminated with the major of isolated fungi i. e. *Aspergillus flavus*, *A. parasiticus*, *Botryodiplodia theobromae*, *Fusarium oxysporum* and *Rhizopus*

stolonifer then incubated at $26\text{ }^{\circ}\text{C} \pm 2$ for two period times i. e. 7 and 14 days. Some physical and bio-chemical characters in both healthy and artificial inoculated Guava fruits were determined in Pomology Dept., National Research Centre (NRC) according to Association of official Agricultural Chemists (A O A C., 2007, Omayma M. Ismail *et al.*, 2010 and Embaby *et al.*, 2012).

1- Physical characteristics

1-1- Fruit weight:

It was determined by weighing the fresh samples weight (between 360-370g) by ordinary balance with 0.01 gm sensitivity and average weight per fruit was calculated compared with healthy (Non-inoculated as a control) and infected once (Omayma M. Ismail *et al.*, 2010).

Loss assessment of Guava fruits was estimated after incubation period in comparison with un-inoculated ones. Percentage of loss was calculated as follows: $\text{Loss} = W_u - W_i$ %Loss = $W_i / W_u \times 100$

%R (%Reduction) = $W_u - W_i / W_u \times 100$ Wheres:

W_u = Weight of un-inoculated fruits

W_i = Weight of inoculated fruits

1-2- Marketable (Shelf life) period after 7 and 14 days stored. Fruit samples from each replicates were stored at room conditions ($26/19\text{ }^{\circ}\text{C}$ and 55-60% RH) till bad appearance or rotting occurs was recorded and considered as shelf life (Omayma M. Ismail *et al.*, 2010).

2-Chemical Characteristics:

2-1 – Total soluble solids content %: Abbe refractometer was used to determine the percentage of total soluble solids content (TSS) in flesh fruit juice from each healthy and diseased fruits. The percentages of TSS were recorded according to (Sharman *et. al.*, 1991, Embaby *et. al.*, 2007 and Omayma M. Ismail *et al.*, 2010).

2-2-Titratable acidity %: Was determined according to the method described in A.O.A.C. (2007). Clear filtrate of inoculated and un-inoculated Guava fruits were used to determine the total titratable acidity (TA) using phenolphthaline as an indicator, after titration with NaOH (0.1 N). The percentage of acidity was calculated as mg citric acid per 100 g fresh weight of

Guava fruit according to the following equation:

Acidity % = $\text{ml of NaOH used} \times \text{N of NaOH (0.1)} \times 0.064 / \text{Sample volume of Guava (ml)}$. Results were expressed as % of malic acid in fresh pulp weight (Omayma M. Ismail *et al.*, 2010).

TSS/Acid Ratio: The total soluble solids (TSS) / total titratable acidity (TA) ratio was calculated directly by dividing TSS value on TA value for each treatment. Ratio = (TSS) / TA

2-3-Ascorbic acid content: Was determined, it was calculated as milligram vitamin C per 100 gm of fresh weight (Embaby *et al.*, 2007 and Omayma M. Ismail *et al.*, 2010). Finally, chemical content losses and reduction percent were calculated as follow:

$$L = H - I \quad \%R = (H - I) / H$$

L = Loss; H=Healthy fruit; I=Infected fruit and %R =Reduction

Results and discussion

Mycoflora associated Guavarotted fruits

During the investigation Guava fruits were found to be susceptible to several fungal diseases, i. e. Aspergillus soft rot (*A. flavus* and *A. parasiticus*), black mould rot (*Aspergillus niger* V. Tiegh.), Botryodiplodiastylar end rot (*Botryodiplodia theobrome*), Fusarium rot (*F. oxysporum*) and Rhizopus soft rot (*R. stolonifethr.* ex Fr.). Healthy and naturally infected symptoms of Guava fruits were photographed (Figs. 1, 2 &3) and the causal agents of fungal pathogens were photographed in Fig. (4.a, b, c, d, e, &f). Analyses of mycoflora associated Guava rotted fruits were recorded in Table (1). Data in this table presented that, one hundred and eighty fungal colonies were isolated from Guava rotted fruits which collected from three different Governorates (Localities), in Egypt. Data also show that, Beheira Governorate (Location) gave higher of total fungal colonies compared with others which record 80 fungal colonies equal 44.44% followed by El-Sharkia Governorate (Location) which record 70 fungal colonies equal 38.89%. Qualubia Governorate (Location) was less fungal colonies and gave only 30 colonies equal 16.67%.

On the other hand data in the same table indicated that, four fungal genera belonging to six species were identified *Aspergillus* (*A. flavus*, *A. niger* and *A. parasiticus*), *Botryodiplodia theobrome*, *Fusarium oxysporum* and *Rhizopus stolonifer*. *Rhizopus stolonifer* was higher fungal frequency occurred which record 81 isolates equal 42.78%, followed by *Aspergillus* (*A. niger* with 48 isolates equal 26.67%, *A. flavus* with 14 isolates equal 7.78% and *A. parasiticus* with 6 isolates equal 3.33%). *Botryodiplodia theobrome* gave 31 isolates equal 17.22 %, while *Fusarium oxysporum* was less fungal frequency occurred which record 4 isolates equal 2.22 %.

Similar results were obtained by many investigators, they reported that, a total of seven (7) fungi were isolated from the postharvest spoilage of Guava

fruit namely *Colletotrichum gloeosporioides*, *Fusarium oxysporum*, *Mucor* sp., *Rhizopus stolonifer*, *Aspergillus niger*, *A. fumigatus* and *A. parasiticus* (Adisa, 1985, Misra, 2004 Ajayi, et al., 2010 and Bhale, 2011). Six fungal organisms were isolated from the samples of guava. The six different fungi viz. *Pestalotia psidii*, *Rhizopus stolonifer*, *Aspergillus niger*, *Penicillium expansum*, *Rhizoctonia solani* and *Fusarium* sp. were found associated with the rotting of the guava fruits Mathew, 2010 and Nonmaithem, 2014.

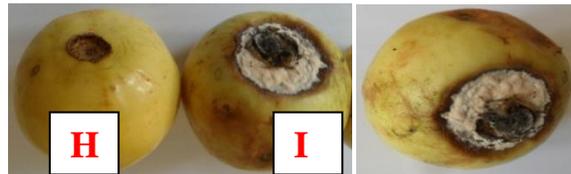


Figure 1. Healthy and naturally infected symptoms of Guava styler end rot disease caused by *Botryodiplodia theobromae* (pre-harvest)



Figure 2. Mechanical longitudinal damage of Guava fruit symptoms, infected with yeast cells and invaded with *Aspergillus niger* (after harvest)

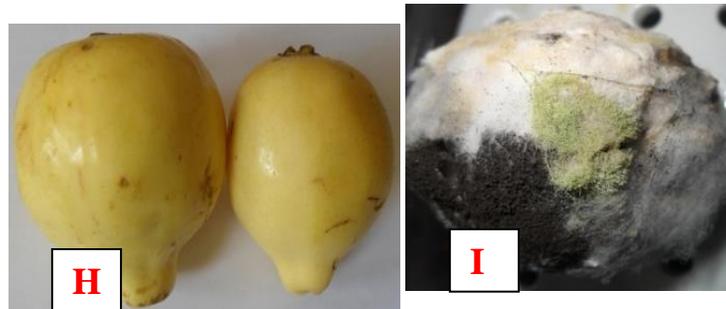


Figure 3. Healthy and infected symptoms with Guava fruit appeared *Aspergillus flavus*, *A. niger*, *Botryodiplodia theobromae* and *Rhizopus stolonifer* symptoms (complete loss during storage)

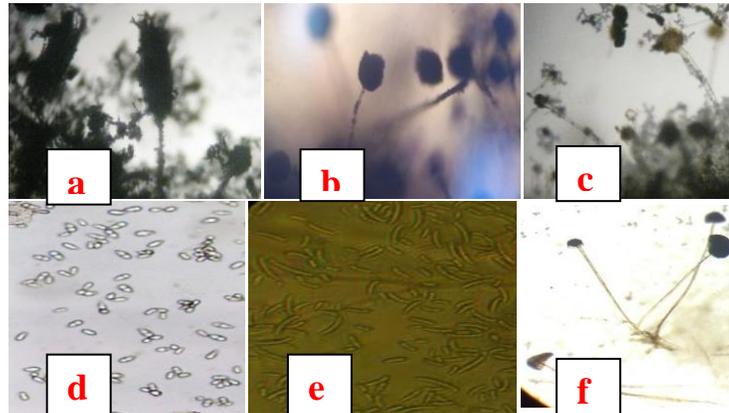


Figure 4. See *Aspergillus flavus*(a), *A. niger*(b), *A. parasiticus*(c), *Botryodiplodia theobromae*(d), *F. oxysporum*(e) and *Rhizopus stolonifer*(f) the causal agent of Guava fruit decay which affecting Guava fruit quality

Table 1. Fungal frequency associated Guava fruit rots

Fungi	Location						Total	
	Beheira		El-Sharkia		Qualubia		T. c	%%
	T. c	%	T. c	%	T. c	%		
<i>A. flavus</i>	6	3.33	5	2.78	3	1.67	14	7.78
<i>A. niger</i>	18	10.00	20	11.11	10	5.56	48	26.67
<i>A. parasiticus</i>	4	2.22	00	0.0	2	1.11	6	3.33
<i>B. theobrome</i>	10	5.56	15	8.33	6	3.33	31	17.22
<i>F. oxysporum</i>	0	0.0	0	0.0	4	2.22	4	2.22
<i>R. stolonifer</i>	42	23.33	30	16.67	5	2.78	81	42.78
Total	80	44.44	70	38.89	30	16.67	180	100.00

T. c= Total colonies

Mycotoxin Determination

Aflatoxins, Ochratoxin A and fumonisin were tested by using high-performance liquid chromatography (HPLC). Data in Table (2) show that, aflatoxins were detected with *Aspergillus parasiticus* only. Aflatoxin G₁ was detected with isolate No. 8 *A. parasiticus* isolated from Qualubia sample. It was less producer of aflatoxin compared with the other which record 0.548 ng/ml. While isolate No. 10, Beheira sample gave higher aflatoxins AFB₁ and AFG₁ which recorded 0.163 ng/ml and 0.296 ng/ml respectively and the total aflatoxins was 0.459 ng/m (Figs. 5 & 6). Neither *A. flavus* nor *A. parasiticus* produced Ochratoxin A. *Fusarium oxysporum* gave negative reaction offumonisin. No mycotoxin was detected with other fungal isolates.

Drusch, and Ragab, (2003) reported that, some potent fungal toxins like aflatoxins, ochratoxinA, patulin have been detected in fruits during storage. Also, Embaby *et al.* (2012) found that, some of moulds could produce mycotoxins while grown on fruits (even during refrigeration). Additionally, if the spoiling fungi are toxigenic or pathogenic, they could pose a health risk for the consumer.

Table 2. Tested of mycotoxin production and its determined
ND = Not detected

<i>Pathogen</i>	<i>Type of Mycotoxins</i>	<i>Beheira</i>	<i>El-Sharkia</i>	<i>Qualubia</i>	
<i>A. flavus</i>	Aflatoxins	AFB ₁	ND	ND	ND
		AFB ₂	ND	ND	ND
		AFG ₁	ND	ND	ND
		AFG ₂	ND	ND	ND
<i>A. parasiticus</i>	Ochratoxin	O A	ND	ND	ND
		Aflatoxins	AFB ₁	0.163 ng/ml	ND
	Aflatoxins	AFB ₂	ND	ND	ND
		AFG ₁	0.296 ng/ml	ND	0.548ng/ml
		AFG ₂	ND	ND	ND
OchratoxinA	O A	ND	ND	ND	
<i>F. oxysporum</i>	Fumonisin	FB ₁	ND	ND	ND
Total		0.459 ng/ml	-	0.548ng/ml	

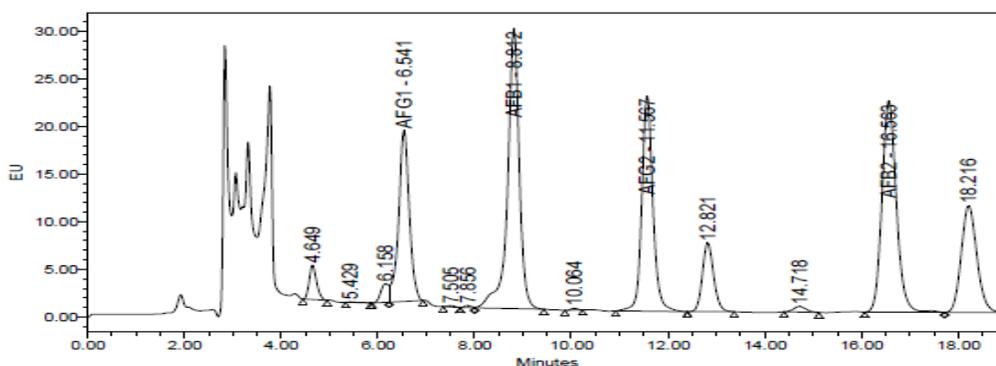


Figure 5. HPLC chromatogram for standard aflatoxins (ST)

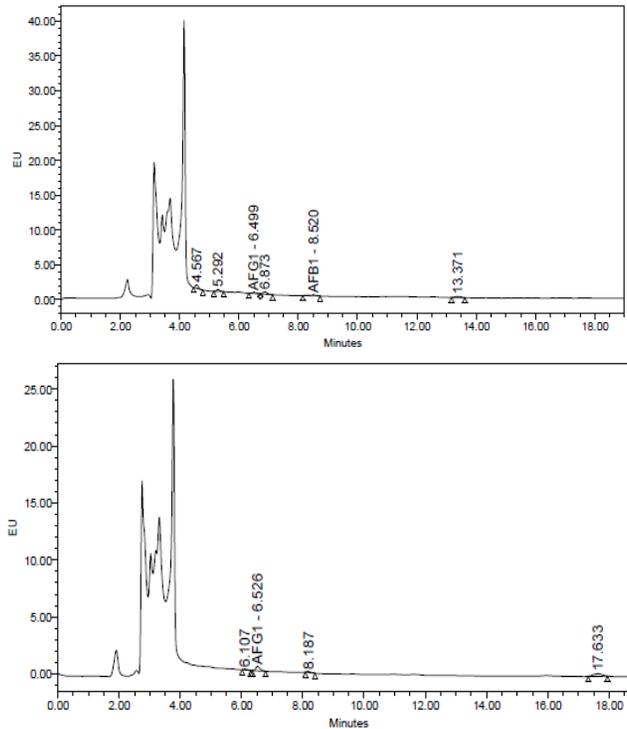


Figure 6. HPLC chromatogram for aflatoxins extracted from *A. parasiticus* isolates No. 8&10 respectively

Decay of Guava fruit quality caused by some mold fungi

Changes in Guava fruit quality caused by the isolated fungi (i. e. *A. flavus*, *A. parasiticus*, *B. theobrome*, *F. oxysporum* and *R. stolonifer*) on some physical and chemical properties and has a limited postharvest shelf lifewhen stored under room condition at 26/19 °Ctemperature with 55-60% relative humidity (RH) after 7 and 14 days were studied and recorded as follow:-

1-Effect ofthe tested fungi on some physical and chemical properties after one weak

Data in **Table (3)** presented that, all tested fungi were found to be decreased all determined of physical and chemical properties copmpared with un-infected Guava fruits.Higher reduction percent with Guava fresh weight was recorded with *R. stolonifer* followed by *B. theobrome*, *F. oxysporum*, *A. parasiticus* and *A. flavus* resectively.Data show that, fresh weight of infected Guava fruits was found to be decreased from 362_(g) with un-infected fruits (control) to 300, 246, 180, 120 and 95_(g) with 17.1, 32.0, 50.3, 66.9 and 73.8

reduction percent after 7 days stored at room condition when infected by *A. flavus*, *A. parasiticus*, *F. oxysporum*, *B. theobrome* and *R. stolonifer* respectively.

The most reduction percent with Vitamin c. content (V. c mg/100 gm flesh) was recorded with *F. oxysporum* followed by both *B. theobrome* and *R. stolonifer*, *A. parasiticus* and *A. flavus* respectively. Vitamin c. content (V. c mg/100 gm flesh) was found to be decreased from 4.0 mg/100 gm in control group to 3.8 mg (5.0% reduction) with *A. flavus fungus*, 3.5 gm (12.5% reduction) with *A. parasiticus*, 3.0 gm (25.0 % reduction) with either *B. theobrome* and *R. stolonifer* in addition 2.2 gm of (V. c) with 45.0 % reduction for *F. oxysporum* fungus. Both *A. parasiticus* and *R. stolonifer* were found to be gave higher reduction percent with total soluble solid followed by either *B. theobrome* and *F. oxysporum* while *A. flavus* was less. *A. flavus* reduced total soluble solid contents from 7% to 6% with 14.3% reduction, both *B. theobrome* and *F. oxysporum* record 5% with 28.6% reduction and 4% with each of *A. parasiticus* and *R. stolonifer* equal 42.9% reduction.

On the other hnd, all tested fungi were found to be increased total titratable acidity compared with un-infected Guava fruits (control group). The percentage of acidity (Total titratable acidity) was found to be increased as mg acid per 100 g fresh weight of Guava fruit from 0.6 mg acid per 100 g with un-infected Guava fruits (control group) to 1.3 mg with *A. flavus*, 1.9 with *A. parasiticus*, 2.5 with *R. stolonifer*, 3.2 with with *B. theobrome* and 3.4 mg with *F. oxysporum*. The total soluble solids (TSS) / total titratable acidity (TA) ratio (TSS/Acid Ratio) was was found to be reduced from 11.7% with un-infected Guava fruits (control group) to 4.6% (60.7% reduction) when infected by *A. flavus*, 2.1% (82.1% reduction) with *A. parasiticus*, 1.6% (86.3% reduction) with both *B. theobrome* and *R. stolonifer* and 1.5% (60.7% reduction) when infected with *R. stolonifer*.

Similar results were obtained by many investigators, they reported that, *Rhizopus sp.* and *A. parasiticus* reduced fresh weight of apricot fruit(s) compared with healthy (un-inoculated). *Aspergillus parasiticus* reduced all chemical contents in all inoculated fruits compared with un-inoculated ones. Higher reduction and loss percent were recorded with total soluble solids (TSS %) followed by total titratable acidity (TA) and ascorbic acid as Vitamin C, while TSS/TA ratio gave the lowest reduction and loss percent. No significant difference in fruits between the TSS/TA ratio percent Embay, et. al., 2007. The most important causal agent responsible for the post-harvest diseases of guava, are the fungi. These microorganisms invade the fruit and cause considerable damage at the post-harvest stage, during transit, storage and transportation to the market Mathew, 2010. Microorganisms also reduce the

quality of the fruit and reduce the percentage of annual production of guava despite all its benefits if not addressed (Ajayi, et al., 2010 and Ammar, and El-Naggar, 2014).

Table 3. Changes in Guava fruit quality in some physical and chemical properties (after 7 days)

Pathogen	Physical properties		Chemical properties						
	W(g)	%R	V. c (mg/100 gm flesh)	%R	(%) Tss	%R	TA	Ratio TSS / TA	%R
<i>A. flavus</i>	300	17.1	3.8	5.0	6	14.3	1.3	4.6	60.7
<i>A. parasiticus</i>	246	32.0	3.5	12.5	4	42.9	1.9	2.1	82.1
<i>B. theobrome</i>	120	66.9	3.0	25.0	5	28.6	3.2	1.6	86.3
<i>F. oxysporum</i>	180	50.3	2.2	45.0	5	28.6	3.4	1.5	87.2
<i>R. stolonifer</i>	95	73.8	3.0	25.0	4	42.9	2.5	1.6	86.3
Control	362	-	4.0	-	7	-	0.6	11.7	-

W (g) = Weight

%R= Reduction

%Tss = Total soluble solids TA = Total titratable acidity

Effect of the tested fungi on some physical and chemical properties (after two weeks)

Changes in Guava fruit quality caused by the isolated fungi (i. e. *A. flavus*, *A. parasiticus*, *B. theobrome*, *F. oxysporum* and *R. stolonifer*) on some physical and chemical properties and has a limited postharvest shelf life after two weeks were recorded in Table (4). Data in this table show that, increasing reduction with all tested fungi with increasing the storage period from one to two weeks which decreased all determined of physical and chemical properties compared with un-infected Guava fruits under room condition.

Data show that, fresh weight of infected Guava fruits was found to be decreased from 356 (g) with un-infected fruits (control) to 162, 160, 131, 112 and 75 (g) with 54.5, 55.1, 62.2, 68.5 and 78.9 reduction percent after 14 days stored at room condition when infected by *A. flavus*, *A. parasiticus*, *F. oxysporum*, *B. theobrome* and *R. stolonifer* respectively. Vitamin c content (V. c mg/100 gm flesh) was found to be decreased from 3.6 mg/100 gm in control group to 3.0mg with 5.0% reduction when infected by *A. flavus* fungus, 2.9gm

(19.4% reduction) with *A. parasiticus*, 2.8gm (22.2% reduction) with *R. stolonifer*, 2.7 with 25.0 % reduction for *B. theobrome* and 1.9 gm of (V. c) with 47.2% reduction for *F. oxysporum* fungus. Also, reduced total soluble solid contents from 7% to 4% (42.9% reduction) with each of *A. flavus*, *B.theobrome* and *F. oxysporum* 3% (57.1% reduction) with *A. parasiticus* and 2% (71.4% reduction) with *R. stolonifer*.

On the other hnd, all tested fungi were found to be increased total titratable acidity compared with un-infected Guava fruits (control group). The percentage of acidity (Total titratable acidity) was found to be increased as mg acid per 100 g fresh weight of Guava fruit from 0.6 mg acid per 100 g with un-infected Guava fruits (control group) to 1.8 mg with *A. flavus*, 2.5 with *R. stolonifer*, 2.8 with *A. parasiticus*, 3.2 with *B. theobrome* and 3.4 mg with *F. oxysporum*. The total soluble solids (TSS) / total titratable acidity (TA) ratio (TSS/Acid Ratio) was found to be reduced from 11.7% with un-infected Guava fruits (control group) to 2.2% (81.2% reduction) when infected by *A. flavus*, 1.3% (88.9% reduction) with *B. theobrome*, 1.2% (89.% reduction) with *F. oxysporum* and 0.8% (93.2% reduction) when infected with *R. stolonifer*.

Embaby *et al.* (2012) reported that, some fungi are plant pathogens and can start the spoilage from the field; they proliferate and cause substantial spoilage only after harvest. Post-harvest fruit spoilage results in significant economic losses.

Table 4. Changes in Guava fruit quality (in some physical and chemical properties (after 14days)

Pathogen	Physical properties		Chemical properties						
	W(g)	%R	V. c (mg/100 gm flesh)	%R	(%) Tss	%R	TA	Ratio TSS / TA	%R
<i>A. flavus</i>	162	54.5	3.0	16.7	4	42.9	1.8	2.2	81.2
<i>A. parasiticus</i>	131	62.2	2.9	19.4	3	57.1	2.8	1.1	90.6
<i>B. theobrome</i>	112	68.5	2.7	25.0	4	42.9	3.2	1.3	88.9
<i>F. oxysporum</i>	160	55.1	1.9	47.2	4	42.9	3.4	1.2	89.7
<i>R. stolonifer</i>	75	78.9	2.8	22.2	2	71.4	2.5	0.8	93.2
Control	356	-	3.6	-	7	-	0.6	11.7	-

W (g) = Weight
titratable acidity

%R =Reduction

%Tss = Total soluble solids
TA = Total

Economic losses and postharvest shelf life after two weeks has been estimated and recorded in Table (5). Data in this table presented that, all tested fungi were found to be decreased Guava shelf life period as well as increased percentage of post-harvest losses compared with un-infected fruits. Higher loss percent of Guava fresh weight (g) was recorded with *A. parasiticus* followed by *A. flavus*, both *F. oxysporum* and *R. stolonifer* respectively while, *B. theobromae* was less. *A. flavus* was the most losses of vitamin C mg/100 gm followed by *A. parasiticus*, *F. oxysporum*, *B. theobromae* and *R. stolonifer* respectively. Higher loss percent of Guava total soluble solids (TSS %) was recorded with *R. stolonifer* followed by *A. flavus*, *A. parasiticus*, both *B. theobromae* and *F. oxysporum* respectively. *A. flavus* was found to be decreased the average of infected fruits from 300g to 162g and lost 138g equal 46.0% reduction, *A. parasiticus* decreased the av. of infected fruit weight from 246g to 131g and lost 115g equal 46.8% reduction, *B. theobromae* decreased the av. of infected fruit weight from 120g to 112g and lost 8g equal 6.7% reduction, both *F. oxysporum* and *R. stolonifer* were found to be decreased the av. of infected fruit weight from 180g to 160g and lost 20g equal 11.1% reduction while, control group (un-infected fruits) was found to be lost only 6g of fruit weight after 14 days equal 1.7% reduction. Vitamin C was found to be decreased from 3.8 to 3.0 mg/100 gm which lost 0.8 mg/100 gm equal 21.1% reduction percent after 14 days when infected by *A. flavus*, from 3.5 to 2.9 mg/100 gm which lost 0.6 mg/100 gm equal 20.7% reduction with *A. parasiticus*, from 3.0 to 2.7 mg/100 gm which lost 0.3 mg/100 gm equal 11.1% reduction with *B. theobromae*, from 2.2 to 1.9 mg/100 gm which lost 0.3 mg/100 gm equal 15.8% reduction with *F. oxysporum* and from 3.0 to 0.2 mg/100 gm which lost 0.2 mg/100 gm equal 7.1% reduction with *R. stolonifer*. Also, *A. flavus* was found to be decreased percentage of total soluble solids (TSS%) from 6 to 4% and lost 2.0/100 ml of juice equal 33.3% reduction, *A. parasiticus* from 4 to 3% and lost 1.0/100 ml of juice equal 25.0% reduction, both *B. theobromae* and *F. oxysporum* were found to be decreased the percentage of total soluble solids (TSS%) from 5 to 4% and lost 1.0/100 ml of juice equal 20.0% reduction and *R. stolonifer* from 4 to 2% and lost 2/100 ml of juice equal 50.0% reduction. While, no changes in total soluble solids (TSS%) with control group (un-infected fruits) after 14 days storage.

Fungi are major spoiling agents responsible for causing post harvest fruit spoilage, leading to significant economic losses Singh and Sharma, (2007). It has been estimated that about 20-25% of the harvested fruits are decayed by pathogens during post-harvest handling even in developed countries (Droby, 2006 and Zhu, 2006). According to a recent newspaper report, annual post-

harvest losses in India are over Rs 2000 billion. There is a considerable gap in food production and net availability to consumers (Embaby *et al.*, 2012 and Vermani *et al.*, 2014).

Guava fruit diseases are of two types, field and postharvest diseases. Postharvest diseases of fruits are the most severe causes of losses in production and are responsible for bio-deterioration of tropical fruit pulp. Storage diseases lead to economic loss by reducing quality and marketability of damaged fruit, or may result in complete loss of the stored fruit (Singh and Sharma, 2007; Nongmaithem, 2014 and Amadi *et al.*, 2014). *Aspergillus parasiticus* reduced significantly fresh weight, fruit quality and all chemical contents. Higher loss percent were recorded with total soluble solids (TSS%), followed by total titratable acidity (TTA) and ascorbic acid (Vitamin C) but TSS/TTA ratio was not significant and showed lowest loss Embaby *et al.* (2007).

Table 5. Percentage of loss and postharvest shelf life after two weeks

Pathogen	Physical properties				Chemical properties							
	W(g)				V. c (mg/100 gm flesh)				(% Tss			
	Period/d.	L	% L	Period/d.	L	% L	Period/d.	L	%			
<i>A. flavus</i>	7	14			7	14			7	14		L
	300	162	138	40.0	3.8	3.0	0.8	21.1	6	4	2.0	33.3
<i>A. parasiticus</i>	246	131	115	46.8	3.5	2.9	0.6	20.7	4	3	1	25.0
<i>B. theobrome</i>	120	112	8	6.7	3.0	2.7	0.3	11.1	5	4	1	20.0
<i>F. oxysporum</i>	180	160	20	11.1	2.2	1.9	0.3	15.8	5	4	1	20.0
<i>R. stolonifer</i>	95	75	20	11.1	3.0	2.8	0.2	7.1	4	2	2	50.0
Control	362	356	6	1.7	4.0	3.6	0.4	11.1	7	7	0	00.0

W (g) = Weight %Tss = Total soluble solids TA = Total titratable acidity %Ratio = TSS /
TA L = Loss % L = % Loss

Conclusion

Fungal infection on Guava fruit may occur during the growing season, harvesting, handling. Storage diseases lead to economic loss by reducing quality and marketability of damaged fruit, or may result in complete loss of the stored fruit. These Fungi invade the fruit and cause considerable damage at the post-harvest stage. Mycotoxins secretion that may be harmful to humans and adept the plant for infection as well as changes in fruit quality.

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