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Bacteriological Analysis of Deteriorated Banana (*Musa Acuminata*) Fruit Sold in Kubwa Market

Maidawa, G.L.; Ayegba, S. O.; Abdullahi, M.; Abdallah, H.Y.; Okoye, C. I.; Idris, H.A.; Adamu, B.B.
National Biotechnology Development agency, Abuja, Nigeria.

Abstract

Fruits are a good source of dietary fibre. They contain vitamins and minerals that keep you healthy. Bananas contain a nutrient that help to manage blood pressure and boosts a person's mood. Bananas are easily damaged and open to infection by bacteria, fungi and viruses. This study was carried out to isolate and identify bacteria associated with spoilt banana. The samples of were collected from Kubwa market in Abuja and were examined by culturing on nutrient, Mac conkey and salmonella-shigella agar. The aerobic mesophilic counts ranged from 2.4 x 10^{-7} cfu/ml to 3.2 x 10^{-6} cfu/ml from the surface wash and the total coliform counts ranged from 2.5 x 10^{7}cfu/ml. fecal coliform ranged from 3.2x 10-6 cfu/ml ad for *Staphylococcus aureus* were 1.8x 10^{-6} cfu/ml and 2.5x 10^{-7} cfu/ml. *Escherichia coli*, *Salmonella sp* and *Staphylococcus aureus* were detected in 89% of the samples. The presence of the isolates may be due to improper hygienic practices from the point of harvesting, transportation and storage by the handlers. Above all the high moisture content of the banana fruit makes it highly perishable as it supports the growth of micro organisms especially bacteria. Therefore it is recommended that the concerned authorities should introduce a supervisory and monitoring system on the sanitary condition of fruits marketing areas and the visibility of transport system.

Keywords: Banana, Isolates, Coliforms, Bacteria, Agar

Introduction

Fruits are very essential and have beneficial standard fruits products have rapidly, expand by more than 30% during the past years. Throughout the interval between 1 870-2004, US per capital utilization of fruits expand.by 19.9% to 694.3 pounds per capita per year. Fresh fruits and legume utiliza6tion expands by

25.8 and 32.6%, sequentially and far better in the rise perceive for refined fruit product (Akinmusire, 2011).

Fruits inhabit a the main character in human diet by providing the necessary evolution and growth factors such as vitamin's and necessary minerals in human daily diet and that can help to retain a standard health. One of the moral factors that control the fruits profitable value is the silent low lead-time phase caused by pathogens strike (Al-Hindi, et al., 2011).

It is expected that about 20-25% of the collected fruits are rotten by pathogens throughout post- harvest handling even in developed countries. If the intrinsic and extrinsic state are suitable, microorganisms build up fast and make delightful food distasteful microbial growth in foods can lead to observable changes, including different colours produced 'by decomposed organisms. A fruit is the consumable part of a grown up ovary of a flowering plant. It is nominally eaten raw. Fruit could also be express as the appetizing of a nut which is mushy, frequently juice is make as they were enlarge from the flower of the plant, they comprise of mature seed or seeds with some tissues attracted (Barth et al., 2009).

Fruits are easily damaged and open to infection by bacteria fungi and viruses. It is approximately one-fourth of the collected fruits are damaged before eating. Microbial damage in fruits range not only with the category, different and structure of fruits. The nature of the damage depends upon the product attacked and the attacking organism. Damage of fresh fruits normally happen during storage and transport. *Osmosphilic microflora* spoil fruits and legume mostly during propagation of their natural acid tolerant and fresh fruits are more responsible to damage for the reasons that watery contents are in connection with breeze and micro-organisms from environments (Droby, 2006).

Microbial invasion of fruits can happen during different stages of its enlargement. Environmental exposure is another factor that contributes to the spoilage for certain time. Due to damage that occurs every year 20% of fruits produce are been lost. Most microorganisms that are originally detected on whole fruits surface are soil inhabitants. Vectors for the distribution of these microbes consist of soil particles, airborne spores, and irrigation water. One of the restrain factors that affect the fruits lucrative value is the proportional short shelf life phase caused by pathogens attacked (Chukwuka et al., 2010).

The utilization of fruits and vegetables helps in guiding human body from number of ailment by providing nutrients, mineral, vitamins and Fibers. Fruits also have different phytochemicals which could have positive influence on the body weight and adjustment and allied situation including diabetes, hypertension as well as to lessen the risk of cancer, cardiovascular disease, cataracts and some of the effective weakness associated with aging Diets that include adequate number of potassium from fruits and

legume also help lessen the chance of having kidney Stones and may help reduce the possessions of boneless. Fruits are also small in calories which assist lower ones calorie intake as part of a weight loss diet (Eckert, et al., 2008).

Banana is an item of edible food also referred to as a berry. It is obtained from flowering plants belonging to the genus Musa. Banana can also be cooked. Banana deterioration could be detected in its colour most times. Microorganisms play a key role in fruit spoilage. The microorganisms include fungi, yeast, bacteria. The deterioration decreases the nutritional and monetary value of the fruits and sometimes may cause health challenges for example gastroenteritis. In growth media contamination may appear due to poor aseptic methods of handling culture. (Schere and Costa, 2010).

Microorganisms associated with the spoilage of banana yeast and mold, eudemons species, micrococcus species, bacterial and fungal species. Looking at many bananas sold in the market today, most of them are infected with either of the above mentioned microorganisms leading to diseases like Soft rot, crown rot bacteria and soft rots which infect -and make these fruits unattractive for human consumption thus posing a health hazard to humans and animals. Hence a need exists to carry out bacteriological analysis on the banana fruits to investigate the major causes of this spoilage and how to control the contamination of banana so that the health of the public would be protected or safe guarded.This study was aimed to carry out bacteriological analysis on spoilt banana sold in Kubwa market in Abuja, F.C.T

Materials and Method

Materials/Equipment

The following hardware materials were used for this research.

i. Banana Fruits
ii. Incubator
iii. Autoclave
iv. Microscope
v. Refrigerator

Media and Reagents used

The following software materials were used for this research.

i. Nutrient Agar
ii. Macconkey Agar
iii. Crystal violet stain
iv. Ethanol
v. Hydrogen peroxide

Sample Collection

In order for the fresh banana fruit to decay, it was stored in a polythene bag after being purchased at the Kubwa Market, F.C.T Abuja.

Preparation of Culture Media

The media (Nutrient and MacConkey agar) for culturing were prepared according to the

manufacturer's directives and autoclaved at 121^{O}C for 15 minutes for 15 lbs pressure. The media were allowed to cool for 45 degrees, then swirled well before aseptically poured into Petri-dishes and allowed to solidify.

Sample Preparation

One grams (1g) of each of the three samples of banana fruit was weighed out using a weighing balance, homogenized into 90 ml of sterile distilled water using a sterile blender respectively.

Laboratory Isolation of Bacteria Organism Associated with spoilt banana

Homogenate samples (Sample 1, 2 and 3) were plated. The plates were swirled clock wisely and anticlockwise, allowed to solidify and then incubated at 37°C for 24 - 48 hours respectively. MacConkey agar, salmonella and shigella agar, and nutrient agar was used for the bacteria isolation. At the end of the incubation periods colonies were expressed as colony forming unit of the suspension. Desired colonies were sub-cultured into fresh agar plates aseptically to obtain pure cultures of the isolates. Pure isolates of the resulting growth were then stored at 40°C.

Biochemical Tests carried for Identification of Bacteria Isolates

Catalase Test

A smear of a small portion of a colony under test was placed into a tube containing about 2ml of hydrogen peroxide. Catalase positive strains caused effervescence (air bubbles) while catalase negative does not.

Oxidase Test

Add 2 -3 drops of Kovac's reagent directly to suspect colonies on an agar plate. Do not flood the plate with the reagent. Observe for color change within 10 seconds. Microorganisms are oxidase positive when the color changes to dark purple within 5 to 10 seconds. Microorganisms are delayed oxidase positive when the color changes to purple within 60 to 90 seconds. Microorganisms are oxidase negative if the color does not change or it takes longer than 2 minutes.

Indole Test

The peptone water medium was inoculated and incubated for 48 hours at 37°C. After 48 hours of incubation, 3 drops of Kovac's reagent was added and was shook very well and allowed to stay for 15 minutes (in each tube). The red ring on the surface of the peptone water indicates positive result, while yellow ring indicates negative result.

Citrate test

A slant plate method is used, inject Simmons citrate agar gently on the slant by slightly touching the tip of the needle to the colony, which is 18 - 24 hours old. In the citrate medium, organisms require more time to grow. Therefore, the solution must be incubated at 35°C to 37°C for 18 - 24 hours. Observe the colour change in the solution, the development of blue colour indicates the alkylation process.

Sugar Fermentation Test

This test was carried out using a media called triple sugar iron agar, containing dextrose (simple sugar), lactose and sucrose. The medium was prepared according to the manufacturer's directive which is to dissolve 65gm into 100ml of distilled water, after which 10ml of the prepared medium was added into test tubes, 3 drops of phenol red was added. Durham's tubes were inserted in an inverted position (for detection of gas production) making sure it touches the medium. The tubes were plugged with non-absorbent cotton-wool and sealed with aluminum foil before being sterilized with autoclave at 105°C for 15 minutes at 151b pressure. After sterilization, each tube were inoculated with specific colony and incubated for 24 hours at 37°C and incubated tubes serve as control.

Acid production was indicated by a change in colour from orange to yellow colour, indicating that acid has been produced and gas production was indicated by the presence of air spaces between the Durham tubes and the medium used presence of air bubbles also indicated presence of gas interring that gas has been produced,

After solidification, the plates were inverted and incubated for 24 hours at 37°C. After incubation, the grown colonies were observed and there morphologies were recorded.

Results

These point bananas were purchase from the local markets in Bida and were observed under different conditions. The spoiled bananas stored in polythene bag was considered as sample 1 and the diluted in sterile saline was considered as sample 2. The one inoculated with sterile nutrient broth for 24 hours was considered as sample 3 the result is presented in the one below.

Table 1: Isolation of bacteria on Different Media

Banana	Nutrient agar	MacConkey's agar	Salmonella and Shigella agar
Sample 1	+	-	+
Sample 2	-	-	-
Sample 3	+	-	+

Key: (+) growth, (-) No growth

Table 2: Biochemical Result of different isolates

Biochemical Test	*E. coli*	*S. aureus*	*S. typhi*
Catalase	+	+	
Oxidase	+	-	-
Indole		+	-
Lactose	A/G	A	A
Glucose	A/G	A	A
Sucrose	A/G	-	-
Xylose	A/G	A	V
Mannitol	A/G	A	A

Key (+) Growth, (-) No growth, A: acid, V; Variable, A/G: Acidic and Gas production
Sample 1 and sample 3 gave positive result on the nutrient agar. MacConkey's Agar, *Salmonella* and *Shigella agar*. Sample 2 did not give any growth on the media used for isolation.

Identification of Organisms

The biochemical tests were performed on the isolate and the required incubation was done at 37^{O}C for 24 hours. The results are displayed in table 2

Aktar et al., (2016) observed the presence of *Echerichia coli* and *Salmonella spp*. All the biochemical test in this study indicates the presence of *Escherichia coli*, *Salmonella typhi* and *Staphylococcus aureus*

Banana (Musa species) yield in an environment which is suitable for the growth of microorganisms most especially bacteria and fungi. This leads to deterioration as a result of microbial activity. .

Deterioration of banana occurs due to some bacterial species causing it to undergo undesirable changes. The results indicate that sample 1 had the highest microbial load when stored and putting bags. While sample 2 did not show any growth and sample 3 also gave positive results from the biochemical results of the study, it was observed that gram-negative bacterial cause banana spoilage. It was also observed that spoilt banana did not indicate the presence of microorganisms when plated immediately without incubation.

Table 3: Number of cfu/ml of Banana

Sample	AMC cfu/ml	TU cfu/ml	FC cfu/ml	Sal cfu/ml	Sta cfu/ml
1	1.82×10^{-7}	1.17×10^{-7}	0.04×10^{-7}	6×10^{-7}	1.62×10^{-7}
2.	0.4×10^{-7}	1.6×10^{-7}	1.5×10^{-7}	0.4×10^{-7}	4×10^{-7}
3.	1.12×10^{-7}	1.09×10^{-7}	0	1.8×10^{-7}	0

Key: AMC: aerobic mesophilic bacteria count T.C: total coliform F.C: Fecal Coliform, Sal: *Salmonella spp*, Sta: *Staphylococcus aureus.*

Table 4: (Mean ±SEM) aerobic mesophilic count, total coliform, fecal coliform, *salmonella spp* and *Staphylococcus aureus* Log 10^1(cfu/ml)

Banana Sample	AMC (Log cfu/ml)	TU (Log cfu/ml)	FC (Log cfu/ml)	Sal (Log cfu/ml)	Sta (Log cfu/ml)
1	2.19^{b}	2.13^{b}	0.76^{b}	0.91^{c}	1.74^{b}
2.	1.82^{a}	1.53^{a}	0.24^{a}	0.61^{a}	1.14^{a}
3.	1.84^{a}	1.85^{a}	1.29^{b}	-0.30^{a}	1.36^{a}

Key: AMC: aerobic mesophilic bacteria, T.C: total coliform, F.C: fecal coliform, Sal: Salmonella spp., Sta: Staphylococcus aureus. Similar letters in columns shoe not significant difference between samples at $p<0.05$.

Discussion

The large bacteria load might be contributed from different sources as pre-harvesting, harvesting and poor handling practices at the post-harvest activities. The assessment on the management of fruit in this market areas indicates that they were different conditions such as poor handling of fruits, sanitary problem of some marketing areas. This condition might have their own contribution on the microbial load for that area.

The difference might be from the handling and sanitary differences in the market. The markets are crowded by vehicles that emits those particles first, the fruits were transported from long distance, harvested on hygienically and third open exposing to several contaminants. When this fruits banana with collected it's maybe exposed to surface from domestic animals which include the source of different contaminants too. Moreover, the fruit sellers put fruits on the ground without covering material. This factor could also increase the microbial load of the banana. There was no statistically significant difference between the main AMC. The market had significant impact on the value of AMC banana supplies. This may be due to different variety variable such

as the handling particle of the banana fruits and sanitary condition of the area (Oyewole, 2012).

The presence of coliform bacteria especially equalized indicates the fashion contamination of the banana fruits which may be true different source of what are you doing pretty harassed and post office activities exposure during transportation facility, in proper storage conditions and for handling practice at any stage and (NACMF, 2018). From statistical analysis p less than 0.05, there was significant difference between Toyota qualifying and facial collision contacts in the market. This may be attributed to sanitary condition of the market area and handling parties 2015.

Conclusion

Banana (Musa species) is a significant step of it important in the developing world microbial storage of fruit may or due to bacteria or fungi causing the fruits to undergo undesirable change. From this study it is concluded that college banana went through that becomes more successively to college causing microorganisms. Inoculation of college banana in nutrients brought also gives presents of microorganisms causing spoilage. Organisms like *Escherichia coli*, *Salmonella spp* stuff areas are present in sport but now causing it the deterioration techniques based on preservation method can be employed to increase this shelf life of the banana.

The presence of these spoilage bacteria poses and economical and health risk to both the farmers and the consumers. The food vendors are on our way of the health risk of fruits that irritate fruits and cause of the duration 4 handling practice during storage measuring, loading, unloading from vehicles inability to use the waste collection being well also identified as definition in the study area markets. When the television fruits are consumed it exposed the consumers to serious health risk and hazard.

The responsibility most especially banana from contamination is shared by everyone involved from the farmer to consumer education and knowledge are tools to improve food safety control. On this basis the following recommendation and stated below

i. Farmers should be trained on the method of fruit harvesting, storage and transportation to markets.
ii. The fruit vendors should be enlightened on how to stop display and sell fruits.
iii. Sensitization of fruits handlers such as food vendors, sellers, workers engaged in loading and unloading fruits from vehicles about fruits bone disease, contamination of fruits, handling of fruits and impact of hygienic problems on the safety of fruit is essential.
iv. Consign cd bodies should introduce supervision and monitoring system on the sanitary condition of fruits marketing areas and the visibility of transport system.

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