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Full Length Research Paper

Isolation and characterization of food spoilage microorganisms at Jimeta by-pass market

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ABSTRACT: Fourteen (14) spoiled food samples were collected at random from the Jimeta by-pass market and characterized microbially and biochemically. Bacterial species such as *Bacillus* species, E. coli, *Klebsiella* species, *Lactobacillus* species, *Micrococcus* species, *Pseudomonas* species, and *Staphylococcus* species were identified, isolated, and characterized using morphological and biochemical reactions. Similarly, morphological characteristics were used to identify, isolate, and characterize fungal species such as *Aspergillus* species, *Fusarium* species, and *Penicillium* species. *Bacillus*, *Klebsiella*, and *Pseudomonas* species were found to be the most prevalent in the spoilage of the majority of the food items. Similarly, as observed in this study, *Penicillium* species, *Aspergillus* species, and *Fusarium* species were arranged in descending order of the frequency of contamination of food items. The rate of contamination of these food items by microorganisms may be linked to unsanitary methods of handling them from harvesting to transportation and storage.

Keywords: Morphological, biochemical, characterization, microorganisms, contamination

INTRODUCTION

Food spoilage occurs during storage and transportation, whether it is cereals, tubers, fruits, or vegetables. Contamination may occur during harvesting, handling, transportation, and/or storage if proper hygiene standards are not met. Food spoilage can be caused by microorganisms in two ways. First, through the active metabolism of food components by living cells. Second, in the absence of live cells, is caused by their intracellular and extracellular enzymes, which react with the food component and change its functional properties, resulting in spoilage.

Food spoilage can occur as a result of physical, chemical, or microbial degradation, with metabolites causing off flavors or textural changes that result in sensory rejection (Chenoll et al., 2005). Microorganisms can contaminate foods at any stage of processing. When food is handled by humans, there is always the possibility of pathogens, air, dust, water, and ingredients adding their share of contamination. Foods come into contact with various people handling the foods between production and consumption. Humans have been a source of pathogenic microorganisms in food, which can cause food-borne diseases, particularly in ready-to-eat fruits and vegetables. Microorganisms in vegetables can originate from a variety of sources, including soil, water, air, animals, and insects. Enteric pathogens can be found in vegetables, especially if animal and human wastes, as well as polluted water, are used for fertilizer and irrigation. Many of these organisms cause various types of product spoilage. Food spoilage occurs when the original nutritional value, texture, and flavor of food are compromised, and the food becomes harmful to people and unfit to eat (Charul et al., 2000).

Fresh vegetables contain microorganisms derived from soil, water, air, and other environmental sources, as well as plant pathogens. Except for tomatoes, which are spoiled not only by yeast and moulds but also by bacteria, the majority of them have high pH (between 5.5 and 6.4). Mould spoilage is caused by *Penicillium, Phytophthora, Alternaria*, *Fusarium, Cladosporium, Trichoderma* and *Aspergillus* species. Among the bacteria, species of *Pseudomonas, Erwinia, Xanthomonas*, Enterobacteria, *Flavobacterium, Lactobacillus, Bacillus* and *Clostridium* are the most important as well as non-faecal enterococci and lactic acid streptococci (Adams and Moss, 1995).

Table 1: Sample type and their codes.

S/NO.	Sample Type	Sample Code
1	Apple	S1
2	Mango	S2
3	Orange	S3
4	Pawpaw	S4
5	Cucumber	S5
6	Tomato	S6
7	Egg	S7
8	Fish	S8
9	Guinea corn	S9
10	Beans	S10
11	Rice	S11
12	Maize	S12
13	Yam	S13
14	Meat (beef)	S14

Characteristics of microorganisms that causes food spoilage

There are three types of microorganisms that cause food spoilage.

Yeasts

Yeast causes fermentation which is the result of yeast metabolism. There are two types of yeasts: true yeast and false yeast. True yeast metabolizes sugar producing alcohol and carbon dioxide gas known as fermentation. False yeast grows as a dry film on surface of a food item such as pickle brine. False yeast occurs in foods that have high sugar or high acid environment (Adams and Moss, 1995).

Moulds

Moulds grow in filaments forming a tough mass which is visible as “mould growth”. Moulds form spores which, when dry, float through the air to find suitable conditions where they can start the growth cycle again (Adams and Moss, 1995). Both yeasts and moulds can thrive in high acid foods at a temperature of 100^{o} C (212^{o} F) in a boiling water canner for the appropriate length of time destroys yeasts and moulds (Adams and Moss, 1995).

Bacteria

Bacteria are round, rod or spiral shaped microorganisms. Bacteria may grow under a wide variety of conditions. There are many types of bacteria that cause spoilage. They can be divided into: spore-forming and nonspore-forming. Bacteria generally prefer low acid foods like vegetables and meat. In order to destroy bacterial spores in a relatively short period of time, low acid foods must be processed for the appropriate length of time at 116^{o} C (240^{O} F) in a pressure canner (temperatures higher than 100^{o} C (212^{O} F) can be obtained only by pressure canning) (Adams and Moss, 1995).

Objective of the study

The main objective of this study is to isolate and characterize food spoilage microorganisms from various food items that are found in Jimeta bypass market.

MATERIALS AND METHODS

The materials collected from Jimeta by-pass market includes Cereals such as guinea corn, maize and rice; Fruits such as banana, orange, pineapple, water-melon; Vegetables such as tomatoes, Irish potato, sweet potato, meat, fish.
These materials were collected in clean sterile polythene bags.

Isolation and characterization of Microorganisms

Isolation of microorganisms was carried out by the method described by Sherman and Cappuccino, (2005). Biochemical characterization of the isolates was done by the methods described by Sherman and Cappuccino, (2005) and Holt *et al*. (1994).

RESULTS AND DISCUSSION

Individual farmers and marketers have suffered serious damage to foods and feedstuffs caused by food-spoilage organisms since time immemorial. They sustain untold losses as a result of insufficient storage facilities, improper use of modern farm machinery, and birds, insect pests, and rodents, all of which contribute significantly to farm produce damage by exposing crops and stored farm products to microbial infection (Okoye, 1991). This study is able to observe the types of microorganisms associated with food spoilage in the Jimeta bypass market, which was established during the construction of the Jimeta Main Market. The market is a mash-up of all the items one would expect to find in a modern market, but the shops and stores where food is

Table 2: Morphological Characterizations of the identified Isolates.

S/NO	Sample code	Isolate code	Colony Morphology	Simple Staining	Gram Staining	Identified Isolate
1	S1	S1 I1	Abundant, opaque, white waxy growth	Rods	Gram +ve rods	*Bacillus* sp.
2	S1	S1 I2	Slimy, white, somewhat translucent, raised growth	Rods	Gram –ve rods	*Klebsiella* sp., *E.coli*
3	S2	S2 I1	Slimy, white, somewhat translucent, raised growth	Rods	Gram –ve rods	*Klebsiella* sp., *E.coli*
4	S2	S2 I2	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli, Klebsiella* sp.
5	S3	S3 I1	Small, creamy, whitish convex colonies.	Rods	Gram-ve rods	*Lactobacillus* sp.
6	S3	S3 I2	Slimy, white, somewhat translucent, raised growth.	Rods	Gram-ve rods	*Klebsiella sp., E.coli*
7	S4	S4 I1	Abundant, opaque, white waxy growth	Rods	Gram+ve rods	*Bacillus* sp.
8	S4	S4 I2	Abundant, opaque, golden growth.	Cocci	Gram+ve cocci	*Staphylococcus* sp., *Micrococcus* sp.
9	S4	S4 I3	Abundant, thin, white growth with medium turning green.	Rods	Gram-ve rods	*Pseudomonas sp.*
10	S5	S5 I1	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli, Klebsiella* sp.
11	S5	S5 I2	Abundant, opaque, white waxy growth	Rods	Gram+ve rods	*Bacillus* sp.
12	S5	S5 I3	Abundant, opaque, golden growth	Cocci	Gram+ve cocci	*Staphylococcus* sp., *Micrococcus* sp.
13	S6	S6 I1	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli, Klebsiella* sp
14	S6	S6 I2	Abundant, opaque, white waxy growth	Rods	Gram+ve rods	*Bacillus* sp.
15	S6	S6 I3	Abundant, opaque, golden growth	Cocci	Gram+ve cocci	*Staphylococcus* sp., *Micrococcus* sp.
16	S6	S6 I4	Abundant, thin, white growth with medium turning green.	Rods	Gram-ve rods	*Pseudomonas* sp.
17	S7	S7 I1	Abundant, thin, white growth with medium turning green	Rods	Gram-ve rods	*Pseudomonas* sp.
18	S7	S7 I2	Abundant, opaque, white waxy growth.	Rods	Gram+ve rods	*Bacillus sp.*
19	S8	S8 I1	Abundant, thin, white growth with medium turning green.	Rods	Gram-ve rods	*Pseudomonas* sp.
20	S8	S8 I2	Abundant, opaque, golden growth	Cocci	Gram+ve cocci	*Micrococcus* sp., *Staphylococcus* sp.
21	S8	S8 I3	Abundant, opaque, white waxy growth	Rods	Gram+ve rods	*Bacillus* sp.
22	S8	S8 I4	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli., Klebsiella* sp.
23	S9	S9 I1	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli., Klebsiella* sp.
24	S9	S9 I2	Abundant, thin, white growth with medium turning green.	Rods	Gram-ve rods	*Pseudomonas* sp.
25	S10	S10 I1	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli., Klebsiella* sp.
26	S10	S10 I2	Abundant, thin, white growth with medium turning green	Rods	Gram-ve rods	*Pseudomonas* sp.
27	S11	S11I1	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli., Klebsiella* sp.
28	S11	S11 I2	Abundant, thin, white growth with medium turning green	Rods	Gram-ve rods	*Pseudomonas* sp.
29	S12	S12 I1	White, moist and glistening growth	Rods	Rods	*E. coli., Klebsiella* sp.
30	S12	S12 I2	Abundant, thin, white growth with medium turning green	Rods	Gram-ve rods	*Pseudomonas* sp.
31	S13	S13 I1	Greenish colonies.	Rods	Gram-ve rods	*Pseudomonas* sp.
32	S13	S13 I2	Cream dull surface with irregular shape	Cocci	Gram+ve cocci	*Bacillus* sp.
33	S14	S14 I1	Abundant, thin, white growth with medium turning green	Rods	Gram-ve rods	*Pseudomonas* sp.
34	S14	S14 I2	White, moist and glistening growth	Rods	Gram-ve rods	*E. coli., Klebsiella* sp.

kept are appalling. As a result, this study was limited to the fourteen (14) food items highlighted in (Table 1). According to the findings of this study, virtually every food item is contaminated by one or more food spoilage organisms. For example, in (Table 2), several food items were discovered to contain more than two isolates, with each isolate contaminated by *Bacillus, Lactobacillus, Streptococcus, Pseudomonas, or Micrococcus* species. The bacterial contamination of food items observed in the Jimeta bypass market is identical, if not worse, in terms of fungal contamination. Occasionally, food items are discovered to be contaminated by more than two fungal organisms. For example, it was discovered that sample S6 (Tomato) was contaminated with *Aspergillus, Fusarium, and Penicillium* species (Tables 3 and 4). The findings of this study demonstrated the prevalence of various food spoilage moulds. Due to the widespread presence of food spoilage molds in this area, some samples were found to be contaminated by multiple molds/fungi. Cereals such as maize, sorghum, and rice, which were staple foods in this region of the country, may be contaminated by a variety of food spoilage molds (Okoye, 1992).

Table 3: Biochemical characterization of the Identified Isolates.

Isolate code	Indole Test	MR Test	VP Test	CU Test	NR Test	Lactose Fermentation	Sucrose Fermentation	Dextrose Fermentation	Identified bacterial isolate
S1 I1	-	-	-	-	+	-	A	A	*Bacillus* sp.
S1 I2	-	+	-	+	+	AG	AG	AG	*Klebsiella* sp.
S2 I1	-	-	-	+	+	AG	AG	AG	*Klebsiella* sp.
S2 I2	+	+	-	-	+	AG	A	AG	*E. coli*
S3 I1	-	-	-	-	+	+	+	+	*Lactobacillus* sp.
S3 I2	-	+	-	+	+	AG	AG	AG	*Klebsiella* sp.
S4 I1	-	-	+	-	+	-	A	A	*Bacillus* sp.
S4I2	-	+	+		+	A	A	A	*Staphylococcus* sp.
S4 I3	-			+	+	-	-	-	*Pseudomonas* sp.
S5 I1	-	+	+	+	+	AG	AG	AG	*Klebsiella* sp.
S5 I2	-	-	+	-	+	-	A	A	*Bacillus* sp.
S5 I3	-	+	-		+	A	A	A	*Staphylococcus* sp.
S6 I1	-	-	-	+	+	AG	AG	AG	*Klebsiella* sp.
S6 I2	-	-	+	-	+	-	A	A	*Bacillus* sp.
S6 I3	-	+	+	-	+	A	A	A	*Staphylococcus* sp.
S6 I4	-	-	-	+	+	-	-	-	*Pseudomonas* sp.
S7 I1	-	-		+	+	-	-	-	*Pseudomonas* sp.
S7 I2	-	-	+	-	+	-	A	A	*Bacillus* sp.
S8 I1				+	+	-	-	-	*Pseudomonas* sp.
S8 I2	-	-	-	-		-	-	-	*Micrococcus* sp.
S8 I3	-	-	+	-	+	-	A	A	*Bacillus* sp.
S8 I4	+	+	-	-	+	AG	A	AG	*E. coli*
S9 I1	+	+	-	-	+	AG	A	AG	*E. coli*
S9 I2	-	-	-	+	+	-	-	-	*Pseudomonas* sp.
S10 I1	-	+	-	+	+	AG	AG	AG	*Klebsiella* sp.
S10 I2	-	-	-	+	+	-	-	-	*Pseudomonas* sp.
S11 I1	-	+		+	+	AG	AG	AG	*Klebsiella* sp.
S1 1 I2	-	-	-	+	+	-	-	-	*Pseudomonas* sp.
S12 I1	-	+	-	+	+	AG	AG	AG	*Klebsiella* sp.
S12 I2	-	-	-	+	+	-	-	-	*Pseudomonas* sp.
S13 I1				+	+	-	-	-	*Pseudomonas* sp.
S13 I2	-	-	+		+	-	A	A	*Bacillus* sp.
S14 I1	-	-	-	+	+	-	-	-	*Pseudomonas* sp.
S14 I2		+	-	+	+	AG	AG	AG	*Klebsiella* sp.

Key: A = Acid; G = Gas; AG = Acid gas; MR =Methyl red; VP = Voges Proskauer; CU = Citrate utilization; NR = Nitrate reduction.

Table 4: Fungal organisms isolated from some of the cereals, fruits and vegetables obtained from Jimeta bypass market.

S/NO.	Sample code	Growth rate	Appearance of colonies	Species
1	S1	1 – 7 days	Green, black	*Aspergillus* spp., *Penicillium* spp.
2	S2	1 – 7 days	White, black, green.	*Candida* spp., *Aspergillus* spp., *Penicillium* spp.
3	S3	1 – 7 days	White, black, green.	*Candida* spp., *Aspergillus* spp., *Penicillium* spp.
4	S4	1 – 7 days	Green, white.	*Penicillium* spp., *Candida* spp.
5	S5	1 – 7 days	Black, green.	*Aspergillus* spp., *Penicillium* spp.
6	S6	1 – 7 days	Black, orange, green	*Aspergillus* spp., *Fusarium* spp. *Penicillium* spp.
7	S8	1 -7 days	Green, black.	*Penicillium* spp., *Aspergillus* spp.
8	S12	1 – 7 days	Orange, black	*Fusarium* spp., *Aspergillus* spp

Key: Sample codes are given in Table 1.

Conclusion

It is evident from the foregoing that several different microorganisms (bacterial as well as fungal) infect or contaminate cereals such as maize and ready- to- eat fruits and vegetables such as oranges, pawpaw and cabbages sold in Jimeta bypass market.

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