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Full Length Research Paper

Microbiology of polyethylene-packaged sliced watermelon (*Citrullus lanatus*) sold by street vendors in Nigeria

E. Nwachukwu*, C. F. Ezeama and B. N. Ezeanya.

*Department of Microbiology, College of Natural and Applied Sciences, Michael Okpara University of Agriculture, Umudike, P. M. B.7267, Umuahia. Nigeria.

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Ten packaged, freshly sliced watermelon were collected from different street vendors to determine their microbiological quality. Eight different microbial isolates were obtained from the sliced watermelon samples, namely *Escherichia coli*, *Klebsiella aerogenes*, *Proteus mirabilis*, *Staphylococcus aureus*, *Lactobacillus* spp., *Saccharomyces cerevisiae, Rhizopus stolonifer* and *Mucor* spp. The effects of high density polyethylene (HDP) and low density polyethylene (LDP) packaging bags on the microbiological quality of freshly sliced watermelon, stored at ambient temperature were also determined. After 10 days of storage, the total viable counts increased from 0.6 x 10^3 cfu/g to 5.3 x 10^3cfu/g and to 5.5 x 10^3 cfu/g in the HDP- and LDP-packaged watermelon samples, respectively. The total fungal counts increased from 0.5 x 10^3cfu/g to 6.7 x 10^3cfu/g and to 7.2 x 10^3cfu/g in the HDP- and LDP-packaged watermelon samples, respectively. Proper sanitation practice and use of good quality packaging materials are recommended in order to avoid risks associated with the consumption of sliced food produce.

Key words: Watermelon, microbiological quality, packaging materials

INTRODUCTION

Over the years, there has been a significant increase in the consumption of sliced produce because they are easily accessible, convenient, nutritious and, most especially, cheaper than the whole fruits or vegetables. This increase in the consumption of sliced fruit has been linked with a parallel increase in food-borne illness (Mensah et al., 1999; Estrada-Garcia et al., 2004). Sliced fruits commonly consumed in Nigeria include paw-paw, pineapple, watermelon and salad vegetables. Water melon is usually produced in the Northern States of Nigeria and brought to the markets from where the street vendors obtain their stock.

Watermelons are usually sliced, packaged in polyethylene bags and sold by street vendors. Their increased consumption, coupled with the associated risk of disease to which consumers may be exposed, is a matter of great concern. It is difficult for one to attest to the hygiene of the processors or to the sanitary conditions at points of preparation. Moreover, the case is worsened by the fact that sliced watermelon street vending is done without adequate storage conditions, thereby exposing the sliced watermelons to flies and other disease-causing agents.

The sliced watermelons are processed and sold by unlicensed vendors with poor education levels and untrained in food hygiene (Muinde and Kuria, 2005; Barro et al., 2006; Barro et al., 2007). The consumption of sliced watermelon may thus potentially increase the risk of food-borne diseases caused by a wide variety of pathogens. There are different sources of microbial invasion of sliced produce. Pathogens may invade the interior surfaces of the produce during peeling, slicing, trimming and other processes like packaging, handling and marketing (Barro et al., 2007). Vendors of street foods usually make use of simple facilities like wheel barrows, trays, mats, tables and make-shift stalls, thus further increasing the risk of food contamination.

Contamination or cross-contamination of street foods, especially sliced fruits and vegetables, are increased by unsanitary processing and preservation methods. The use of dirty utensils, as well as the open display of street food produce encourages sporadic visits by flies, cock

*Corresponding author. E-mail; drejik@yahoo.com.

Table 1. Total viable counts (cfu/g) of street vended watermelon samples.

Samples*	Bacterial count	Fungal count
A	2.3×10^5	2.8×10^5
B	1.8×10^5	2.8×10^5
C	0.3×10^5	0.4×10^5
D	0.1×10^5	0.2×10^5
E	0.4×10^5	0.9×10^5
F	1.6×10^5	2.4×10^5
G	1.7×10^5	1.9×10^5
H	0.3×10^5	0.9×10^5
I	0.5×10^5	0.3×10^5
J	1.9×10^5	1.8×10^5

* Sliced watermelons (A to J) were collected randomly from different street vendors.

cockroaches, rodents and dusts (Bryan et al., 1992). Preservation of sliced fruits and vegetables that requires no further processing before consumption at ambient temperatures during retail maintains the produce at optimum temperatures for invasion by pathogenic mesophiles (Muinde and Kuria, 2005;

Another major source of contamination of fresh fruits and vegetables sold by street vendors is the washing water (Khali et al., 1994). Poorly processed street vended produce have been identified as an important cause of deaths in developing countries (Mensah et al., 2002). Bacteria like *Salmonella* spp., *Shigella* spp., *Campylobacter* spp. and *Escherichia coli* can contaminate sliced fruits and vegetables through contact with sewage and contaminated water (Fredlund et al., 1987; Blostein, 1991; Beuchat, 1995; Gayler et al., 1995).

The aim of this study was to determine the microbiological quality of sliced watermelon sold by street vendors and to investigate the effects of packaging materials, such as high and low density polyethylene bags, on the microbiological quality of stored sliced watermelon.

MATERIALS AND METHODS

Watermelon samples and culture media

Ten different samples of packaged, sliced watermelon were randomly purchased from ten different street vendors in Owerri metropolis and placed in plastic sterile bags. The samples were immediately transported in a cool container to the laboratory and analyzed within 1 - 2 h after collection. MacConkey agar (Antec Diagnostics Products, UK), Nutrient agar and Sabouraund Dextrose agar (International Diagnostics Group, UK) were prepared and used for the isolation and enumeration of bacteria and fungi. The various media were prepared, as specified in the manufacturer's manuals.

Isolation and enumeration of bacteria and fungi

A sterile knife was used to cut 1.0 g from each watermelon sample and it was then homogenized using a sterile mortar and pestle. The resultant homogenate was added to 9.0 ml of distilled water in a test tube and diluted serially. From the appropriate dilution, 0.1 ml was plated onto the different media. The MacConkey agar and Nutrient agar plates were incubated at 37°C for 48 h to obtain the total viable bacterial counts, while the Sabouraund Dextrose agar plates were incubated at 28°C for 72 h to obtain the fungal counts. These experiments were repeated on three different occasions for each of the samples and the average values recorded. Discrete colonies were streaked onto fresh agar to obtain pure cultures of the different isolates. Isolates were maintained on Nutrient agar slants and stored at 4°C for further tests. Biochemical tests, performed on the bacterial isolates included Gram staining, catalase activity, sugar utilization, methyl red and Voges Proskauer tests coagulase activity, citrate utilization and motility (Baker and Breach, 1980). Fungal identification and classification were based on their macroscopic and microscopic features. The macroscopic features were based on the shape, colour and physical appearance of the colonies, while the microscopic characterizations were carried out according to the methods and identification keys of Fawole and Oso (1986).

Comparison of different polyethylene bags as packing material for sliced watermelons

A watermelon sample was cut with a sterile knife into 15 pieces of 1.0 g each. As a control, five pieces of watermelon were placed on a clean plate, and left uncovered at ambient conditions. Five watermelon pieces were each wrapped separately in either low density (20 µm thick) or high density (12 µm thick) polyethylene bags. The bags were placed on a clean plate and stored at ambient conditions. Total viable bacterial and fungal counts were determined, as described in the preceding section, after one, three, five, seven and ten days of storage.

Statistical analysis

The data obtained was subjected to analysis of variance for mean comparison of different packages at the probability level of $p = 0.05$.

RESULTS

Microbiological analysis of street vended sliced watermelon

The total bacterial and fungal counts were determined for each of the 10 sliced watermelons that were collected randomly from different street vendors, the results of which are shown in Table 1. The total viable bacterial count ranged between $0.1 - 2.3 \times 10^5$ cfu/g, and the total fungal count was similar to that above, ranging between $0.2 - 2.8 \times 10^5$ cfu/g. A total of 40 microbial isolates, comprising both bacteria and fungi, were obtained from 10 sliced watermelon samples analyzed in the present study (Table 1). The bacterial isolates were identified as *E. coli*, *Klebsiella aerogenes, Proteus mirabilis*, *Staphylococcus aureus* and *Lactobacillus* spp., while the fungal isolates were identified as *Rhizopus stolonifer*, *Saccharomyces cerevisiae* and *Mucor* spp. Amongst the bacterial isolates, both *E. coli* and *K. aerogenes* were each isolated from 6 of the 10 sliced watermelon samples analyzed, followed by *P. mirabilis* (4 watermelon samples). In contrast, *S. aureus* (2 samples) and *Lactobacillus* spp. (1

Table 2. Occurrence of bacterial and fungal isolates in street vended sliced watermelon samples.

Sample	*Escherichia coli*	*Klebsiella aerogenes*	*Proteus mirabilis*	*Staphylococcus aureus*	*Lactobacillus* spp.	*Rhizopus stolonifer*	*Saccharomyces cerevisiae*	*Mucor* spp.
A	+	+	–	–	–	+	+	–
B	–	–	–	+	–	+	+	–
C	+	–	+	–	–	+	–	+
D	+	–	–	–	–	+	–	+
E	–	+	–	–	–	+	+	–
F	+	+	–	–	+	+	+	+
G	+	–	+	–	–	+	+	–
H	–	+	–	–	–	+	+	–
I	–	+	+	–	–		+	–
J	+	+	+	+	–	+	+	+

Table 3. Total viable counts (x 10 ^{3} cfu/g) of bacteria and fungi from sliced watermelon packaged in different polyethylene bags

Microbe	**Day 1**			**Day 3**			**Day 5**			**Day 7**			**Day 10**		
	HDP*	LDP#	CNT§	HDP	LDP	CNT	HDP	LDP	CNT	HDP	LDP	CNT	HDP	LDP	CNT
Bacteria	0.6	0.6	0.6	1.1	1.8	2.1	1.4	2.0	3.8	3.4	3.5	8.0	5.3	5.5	TNTC†
Fungi	0.5	0.5	0.5	1.7	2.2	3.9	3.3	3.9	7.0	4.9	5.8	10.9	6.7	7.9	TNTC

HDP* High density polyethylene bag
LDP# Low density polyethylene bag
CNT§ Control
TNTC† Too Numerous To Count

sample) were isolated infrequently. Amongst fungi, *R. stolonifer* was isolated the most frequently (9 samples), followed by *Mucor* spp. (4 samples), while the yeast *S. cerevisiae* was isolated from 8 watermelon samples.

Evaluation of different polyethylene bags as a means to store sliced watermelons

Results of total viable bacterial and fungal counts performed on sliced watermelon samples wrapped in high density and low density polyethylene bags, respectively, are shown in Table 3. As expected, both the total viable bacterial and fungal counts of the control watermelon slice, which was left unwrapped and exposed to the environment, increased over time and on the tenth day, the microbial populations were too numerous to count. The total viable bacterial and fungal counts of sliced watermelon samples that were wrapped in the high density and low density polyethylene bags also showed an increase in counts over time. The total viable bacterial counts increased from 0.6×10^3 cfu/g (day 1) to 5.3×10^3 cfu/g (day 10) in the samples wrapped in high density polyethylene (HDP) bags, and to 5.5×10^3 cfu/g (day 10) in the samples wrapped in low density polyethylene (LDP) bags. The fungal counts increased from 0.5×10^3 cfu/g (day 1) to 6.7×10^3 cfu/g (day 10) in the samples wrapped in HDP bags and to 7.2×10^3 cfu/g (day 10) in the samples wrapped in LDP bags (Table 3).

DISCUSSION

Different microorganisms were isolated from sliced watermelon sold by street vendors (Table 2). The presence of these organisms can be linked to a number of factors such as improper handling and processing, use of contaminated water during washing, cross contamination from other fruits and vegetables or the use of dirty processing utensils like knives and trays (Bryan et al., 1992; Khali et al., 1994).

The high frequency of isolation of *E. coli* and *K. aerogenes*, which are faecal and non-faecal indicators of water pollution, respectively, implicated the processing and rinsing water as possible sources of contamination by these bacteria of sliced watermelons sold by street vendors. The presence of *Staphylococcus* may be explained by the fact that human beings, that is, processors or vendors, carry these organisms on/in several parts of their bodies (Nester et al., 2001). This can be introduced into the fresh sliced watermelon during handling, processing or vending. The low levels of isolation of *Lactobacillus* spp. and *S. aureus* in fresh sliced water-melon is surprising. Generally, *Lactobacillus* spp. belongs to the group of lactic acid bacteria and thrives well in an acid medium, while *Staphylococcus* spp. is salt-loving bacteria (Nester et al., 2001). This, therefore illustrates the need for adequate evaluation of the physicochemical (including pH) characteristics of the watermelon. The presence of *S.*

cerevisiae, R. stolonifer and *Mucor* spp. is in agreement with the report of Splittstoesser (1987) who implicated fungi as contaminants of fresh fruits, especially in the presence of injuries like slicing. Water and the environment may have played major role in fungal contamination of the watermelon samples, especially during washing of the fruits. The high occurrence of *R. stolonifer* could be due to the marked sporulating ability; hence the ease to contaminate the atmosphere, water and fruits. The rather low occurrence of *Mucor* spp. may have been due to the fact that the species require organic matter for growth. The organisms isolated may cause diseases that vary in severity from mild gastroenteritis to severe and sometimes chronic or opportunistic infections.

Studies on the use of high density polyethylene (HDP) and low density polyethylene (LDP) bags for packaging of sliced watermelon showed that bacteria and fungi can survive in both packages when stored at ambient temperatures. The differences in the microbial numbers of watermelon in HDP or LDP bags and the unpackaged watermelon samples were statistically significant ($p<0.05$). However, the (HDP)-packaged samples have the least bacterial and fungal counts compared to the (LDP)-packaged samples and the control experiment. The possible reason for the difference may be attributed to atmospheric conditions. Packaging alters the gaseous composition of the micro-environment and thus results in differences in bacterial and fungal growth. The packages recorded higher fungal counts as compared with bacterial counts. Similarly, Efiuvwevwere and Oyelade (1991) reported that packed oranges in HDP or LDP bags were more susceptible to fungal spoilage.

The result of this study has shed light on the microbial quality, as well as the effect of package materials on the microbiological quality of stored sliced watermelons sold by street vendors. In order to minimize the contamination level of watermelons, good hygiene and the use of HDP packaging materials are required. However, proper sanitary condition must be practiced by food vendors. In addition, the government must have a better surveillance on the activities of sliced produce street vendors in order to minimize the risk of disease outbreak associated with consumption of contaminated food produce.

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