



MICROBIOLOGICAL ASSESSMENT OF *CAPSICUM SPP.* SOLD WITHIN A RURAL MARKET IN DELTA STATE, NIGERIA

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Abstract

Microbiologically safe vegetables are essential to maximize the health benefits within by adequate consumption of these produce. Proper washing of vegetables is essential for decontamination of plant and human pathogens associated with them. To assess the microbial quality of some Capsicum spp. sold in Amai local market, various fresh and dry samples were obtained from two vendors. Samples were analyzed to determine the density of microorganisms by Standard Plate Count, the characterization and identification using biochemical and fungal tests were conducted. Mean bacterial load (fresh and dry samples) ranged from $4.0 \times 10^6 - 6.5 \times 10^7$ cfu/ml for vendor A and $0.5 \times 10^6 - 7.0 \times 10^7$ cfu/ml for vendor B while the fungal load (fresh and dry samples) for vendor A ranged from $3.2 \times 10^6 - 8.6 \times 10^7$ cfu/ml and vendor B ranged from $6.4 \times 10^6 - 8.0 \times 10^7$ cfu/ml. The bacteria isolated with percentage occurrence include: Bacillus spp. (44.4), Serratia marcescens (11.1), Micrococcus spp. (22.2), Enterobacter aerogenes (11.1), S. aureus (11.1) while fungi include: Aspergillus niger (33.3), Penicillium spp. (33.3) and Yeast (33.3). The high isolation rates observed in this study indicate heavy microbial contamination of the vegetable under study, which could result from the cultivating, harvesting or post harvest processing. It is of paramount importance that effective treatment or processing be embarked upon to safeguard consumers' health in the consumption of raw or cooked produce.

Keywords:

Microbiological assessment, *Capsicum spp.*, food borne pathogens, contamination

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1.0 Introduction

Fruits and vegetables are an extraordinary dietary source of nutrients, micronutrients, vitamins and fibre for humans and thus vital for health and well being. Well balanced diets, rich in fruits and vegetables are especially valuable for their ability to prevent vitamin C and vitamin A deficiencies and are also reported to reduce the risk of several diseases [1].

Fruits and vegetables are widely exposed to microbial contamination through contact with soil, dust and water and by handling during cultivation, harvesting, transporting, packaging, storage and selling to the final consumer. They therefore harbor a diverse range of microorganisms including plant and human pathogens [2]. Microbial spoilage and contaminating pathogens pose a serious problem in food safety. Differences in microbial profiles of various fruits and vegetables result largely from unrelated factors such as resident microflora in the soil, application of non-resident microflora via animal manures, sewage or irrigation water, transportation and handling by individual retailers

[3][4]. In developing countries such as Nigeria, continued use of untreated waste and manure as fertilizers for the production of fruits and vegetables is a major contributing factor to contamination [5][6].

Despite their nutritional and health benefits, outbreaks of human infections associated with the consumption of fresh or minimally processed fruits and vegetables have increased in recent years [7]. Enteric pathogens such as *E. coli* and *Salmonella* are among the greatest concerns during food-related outbreaks [8]. Several cases of typhoid fever outbreak have been associated with eating contaminated vegetables grown in or fertilized with contaminated soil or sewage [9]. These increases in fruits and vegetables-borne infections may have resulted from increased consumption of contaminated fruits and vegetables outside the home as most people spend long hours outside the home [2]. The lack of an effective antimicrobial treatment at any step from planting to consumption and post harvest processing equipments of raw farm produce means that pathogens introduced at any point may be present on

the final product. However, fresh vegetables and fruits must be washed or treated specifically to minimize microbial load. This study, therefore intends to assess the microbial load of pepper (*Capsicum spp.*) cultivated, harvested and sold within a local market, which invariably is transported to the urban market for retailing and consumption.

2.0 MATERIALS AND METHODS

2.1 Sample Collection

Fresh and dry samples of *Capsicum annuum* (Red bell pepper) and *C. frutescens* (African bird's eye) were collected from two main wholesale vendors in Amai local market in Delta State, Nigeria, in sterile plastic bags and transported to the laboratory for processing. All analysis was carried out at the Biological sciences department, Novena University, Ogame, Delta state.

2.2 Isolation and Identification of microorganisms

Pour plate techniques were carried out [10]. 25g of each sample was weighed, macerated and washed in 100ml of sterile distilled water. 1ml of the rinse water was inoculated into already prepared molten Nutrient agar (NA), MacConkey agar (MA), and Sabouraud-dextrose agar (SDA). The NA and MA plates were allowed to solidify, inverted and incubated at 37 °C for 24 h while SDA plates were incubated at room temperature for 48h for colony formation. Distinct colonies on plates were sub-cultured on freshly prepared MA and SDA plates and incubated as above. Distinctive morphological properties of each pure culture such as colony formation, elevation of colony, pigmentation and colony margin were observed. Further identification and characterization were carried out using Gram staining, motility, catalase, coagulase, oxidase, lactose utilization, indole test, citrate utilization, urease, sugar

fermentation, MR-VP test for bacterial isolates and cotton lactose-phenol blue test for fungi isolates.

2.3 Enumeration of Microbial load

Enumeration of microorganisms present in each sample were determined by using 10-fold serial dilutions of each rinse water and 1ml of 10^{-2} , 10^{-4} and 10^{-5} dilutions were pipette into sterile Petri dishes and molten Nutrient agar was added and swirled thoroughly to allow even distribution. The colonies were recorded as colony forming units per gram (cfu/g).

3.0 RESULTS

All fresh and dry samples examined in this study were contaminated with varying microbial loads from the two vendors (Table 1). Bacterial load (fresh and dry samples) ranged from $4.0 \times 10^6 - 6.5 \times 10^7$ cfu/ml for vendor A and $0.5 \times 10^6 - 7.0 \times 10^7$ cfu/ml for vendor B while the fungal load (fresh and dry samples) for vendor A ranged from $3.2 \times 10^6 - 8.6 \times 10^7$ cfu/ml and vendor B ranged from $6.4 \times 10^6 - 8.0 \times 10^7$ cfu/ml. The bacterial isolates obtained from fresh samples of *C. annuum* and *C. frutescens* with their percentage occurrence included: *Bacillus spp.* (44.4), *Serratia marcescens* (11.1), *Micrococcus spp.* (22.2), *Enterobacter aerogenes* (11.1), *S. aureus* (11.1) while the fungal isolates included: *Aspergillus niger* (33.3), *Penicillium spp.* (33.3), Yeast (33.3) (Table 2). *Bacillus spp.* was the only bacterial isolates obtained from the dry samples of *C. annuum* and *C. frutescens* while the fungal isolates were as recorded for fresh samples. The non-isolation of other bacterial isolates in dry samples could be as a result of low water activity (a_w) in sample and the antimicrobial properties of capsaicin and dihydrocapsaicin present in the *Capsicum spp.*

Table 1. Total plate count of *Capsicum annuum* and *C. frutescens* obtained from Amai local market, Delta State

Sample	Microbial load (cfu/ml)			
	Average bacterial count		Average fungal count	
	vendor A	vendor B	vendor A	Avendor B
Fresh <i>C. annuum</i>	4.7×10^6	3.3×10^6	8.6×10^6	8.0×10^7
Dry <i>C. annuum</i>	4.0×10^6	0.5×10^6	3.2×10^6	2.5×10^7
Fresh <i>C. frutescens</i>	6.5×10^7	7.0×10^7	8.6×10^7	6.4×10^6
Dry <i>C. frutescens</i>	2.5×10^7	4.0×10^7	3.0×10^6	2.0×10^7

Table 2. Bacterial and Fungal isolates with percentage occurrence in *Capsicum annum* and *C. frutescens* from Amai local market

Isolates	Fresh <i>Capsicum spp.</i>	Freq.	Samples			
			Dry <i>Capsicum spp.</i>		Freq. no (%)	
			no	no		
Bacterial	<i>Bacillus spp.</i>		3	<i>Bacillus spp.</i>	1	4(44.4)
	<i>Serratia marcescens</i>		1	-	-	1(11.1)
	<i>Micrococcus spp.</i>		2	-	-	2(22.2)
	<i>Enterobacter aerogenes</i>		1	-	-	1(11.1)
	<i>Staphylococcus aureus</i>		1	-	-	1(11.1)
						<u>9(99.9)</u>
Fungal	<i>Aspergillus niger</i>		1	<i>Aspergillus niger</i>	1	2(33.3)
	<i>Penicillium spp.</i>		1	<i>Penicillium spp.</i>	1	2(33.3)
	<i>Yeast</i>		1	<i>Yeast</i>	1	2(33.3)
						<u>6(99.9)</u>

Discussion

Microorganisms which are generally ubiquitous have been demonstrated in the fresh and dry samples of *C. annum* and *C. frutescens*. Since moisture is one of the physical factors that facilitate growth of microorganisms, the frequency of isolation is higher in fresh pepper samples than dry samples. This result was in agreement with an earlier study [11] on the microflora of black and white pepper. The presence of these organisms are a direct reflection of the poor sanitary condition of the cultivation water, harvesting, transportation, storage and processing of the produce [2][3].

The bacteria and fungi isolated in this study have previously been isolated from fruits and vegetables (*Capsicum annum*) in other studies both within Nigeria and elsewhere [2][12][12]. The high occurrence of bacteria and fungi (10^6 to 10^7 cfu/ml) in this study was in consonance with earlier studies [2][14][12]. The presence of *S. aureus*, a pathogenic organism of public health importance in most samples under study and the presence of *Bacillus spp.*, agent of food-borne infection and other food quality indicator like *Enterobacter aerogenes* and *Micrococcus spp.*, further reveal the need for proper washing, decontamination and effective processing of the food spices under study, which will enhance the safeguard of consumers health (both at the local and urban areas) before consumption of the raw or processed products. This study thus gives a general overview of the microbiological quality of fresh and dry *C. annum* and *C. frutescens* sold within a local market in Delta State, Nigeria.

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