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Sanitary quality and proximate composition of vernonia amygdalina, telfairae occidentalis and talinum triangulare commonly used for Nigerian dishes

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Abstract

Vegetables are an important part of human diet because of its nutritious contents and roles it plays in fighting diseases away from our body. 30 vegetable samples comprising of Vernonia amygdalina (bitter leaf), Telfairae occidentalis (pumpkin leaf), and Talinum triangulare (waterleaf) were purchased randomly from the Choba market so as to evaluate the microbial quality using standard microbiological methods. The total heterotrophic bacterial count of these leafy vegetables ranged from $5.45x10^6$ to $2.57x10^7$cfu/g. The percentage occurrence of the bacteria species isolated from the vegetable samples are Escherichia coli 25%, Salmonella sp 15.59%, Staphylococcus sp 21.15%, Bacillus sp 11.51%, Klebsiella sp 7.69%, Shigella sp 7.69%, Micrococcus sp 5.77% and Enterobacter sp 5.77%. The fungal counts of the vegetable samples ranged from $1.5x10^4$ to $7.1x10^4$cfu/g. The percentage occurrence of the fungal species isolated are Aspergillus sp 40%, Candida sp 20%, Pencillum sp 20%, Fusarium sp 6.67%, and Geotrichum sp 13.33%. It was observed that microbial contamination of various vegetable samples are as a result of indigenous microbes that are found in the soil and poor hygienic practices of vendors. Both the vendors and consumers should be enlightened on the sanitary practices while displaying, selling and processing of vegetables and health implications of these pathogenic organisms associated with vegetables so as to avoid foodborne infections.

Keywords: Sanitary, vernonia, talinum.

Introduction

Vegetables can be eating by both humans and animals as food. Vegetables are generally regarded as all edible parts of a plant which include its flowers, fruits, stems, leaves, roots, and seeds. Vegetables are grown all over the world mostly in conducive environments. The environmental condition of a place or country is dependent on the type of fruits and vegetables that can be found in such places. Vegetables can be cooked or taken raw and it helps to build our immunity, because of its low carbohydrate and fat content. Vegetables are rich in vitamins, minerals, and dietary fiber. Many people are often encouraged to eat plenty of fruits and vegetables because of the roles it plays in human health. Fresh green vegetables have been linked to food borne diseases in humans worldwide, particularly this has occurred because fresh green vegetables or its ready-to-eat salad have been taken raw without minimal treatment or proper cooking[1].

There is a drastic increase in the outbreak of diseases associated with consumption of fresh food in the United States as reported by the centre for diseases control and prevention (CDC) (1995-2005)[2]. Contamination of fresh fruits and vegetables are as a result of several factors used in farming practice by the farmer while growing his produce. Some of these factors include; polluted irrigation water, soil and unhealthy seed, surrounding animal manure, and poor hygienic practices of workers[3]. Elderly people, children, and immunosuppressed patients stand at a high risk of contracting pathogens, which might harbor in contaminated fruits and vegetables. Microorganisms found in various fruits and vegetables differs mostly as a result of indigenous organisms found in the soil, introduction of non-indigenous organisms through animal manure, sewage, or polluted irrigation water, transportation, and handling by the retailers[4,5].

Contamination of fresh fruits and vegetables in Nigeria, is as a result of polluted water and untreated sewage used while farming[6,7]. Consumption of fresh fruits and vegetables has been linked to various food-borne diseases in recent years. Food-borne diseases is a general problem all over the world.

The health and productivity of the population of most countries, especially non-developed ones have been seriously affected due to the incidence of food-borne diseases[8]. Deterioration of vegetables are mostly caused by the following bacteria; Bacillus cereus, Micrococcus sp., Staphylococcus aureus, Staphylococcus epidermidis, Escherichia coli, Proteus mirabilis and Enterobacter aerogenes[9].

Materials and methods

Area of study: 30 vegetable samples comprising of bitter leaf, pumpkin leaf and water leaf were gotten from Choba market.

Sample collection: A total of 30 different vegetable samples were purchased randomly at Choba market, and was taken to the laboratory for analysis within an hour of collection. The samples were properly labeled according to how they were purchased from the market. The first sample was bitter leaf (Vernonia amygdalina). The second sample was pumpkin leaf (Telfaira occidentalis). The third sample was waterleaf (Talinum triangulare).

Microbial analysis of the vegetables: 25g of each of the samples (non-homogenized leaves) were measured and placed into 250ml of peptone water in a conical flask. The leaves were stirred gently using a sterile glass rod for about 5 minutes. From the stock solution, 1ml was added to 9ml of sterile diluent in the test tube to obtain a dilution factor of 10^{-2}, 1ml was added to the second test tube from the first to obtain a dilution factor of 10^{-3}. A dilution factor of 10^{-6} was obtained from the process. 0.1ml of the solution from the test tubes with a dilution factor of 10^{-4} and 10^{5} were dropped on the general purpose media. 0.1ml of the solutions with a dilution factor of 10^{-2} and 10^{-3} were dispensed on the selective media. The aliquot, with dilution factors of 10^{-4} and 10^{-5} was placed on the general purpose media and was used for total viable heterotrophic count. On mannitol salt agar, 0.1ml from the dilution factor of 10^{-2} and 10^{-3} were used for Staphylococcus sp. count. Coliform count was on MacConkey agar. Salmonella sp. and Shigella sp. were isolated on Salmonella-Shigella media. Potato dextrose agar was used to isolate the fungi species[10].

Examination of fungi: Identification of fungi species are based on their physical characteristics such as colour and cell morphology. Light microscope is used for the microscopic morphology of the fungi species ones properly stained with lactophenol blue on a glass slide.

Proximate analysis of the vegetables: This was done as described by Ray, B. and Bhunia, A.K.[11].

Results and discussion

Table-1: Total means value of bacteria count, staphylococcus count and coliform count of different vegetable samples.

Sample Vegetables	Mean value of heterotrophic bacteria count	Mean value of staphylococcus count	Mean value of coliform count
B	7.08	5.3	4.58
P	7.32	5.39	4.64
W	7.12	5.32	4.44

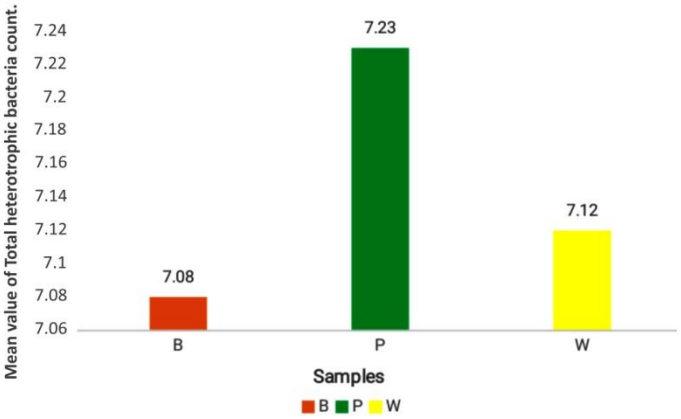

Figure-1: Total Mean of Total Heterotrophic Bacteria Count of the Different vegetables KEYS B = Bitter leaf, P = Pumpkin leaf W = Water leaf.

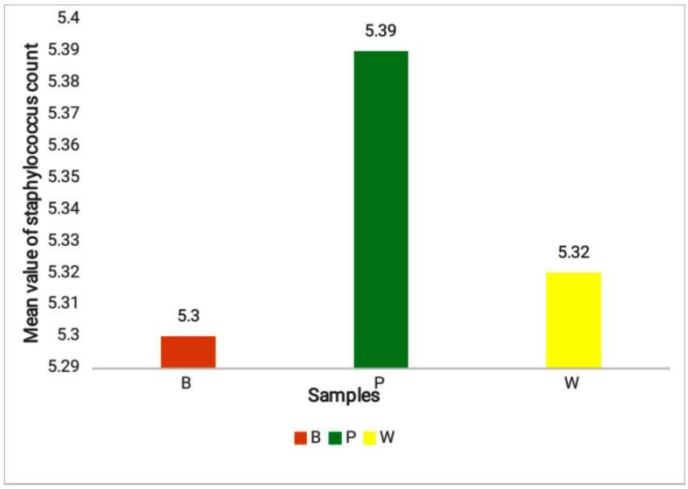

Figure-2: Total Mean of Total Staphylococcus Count of the Different vegetable KEYS B = Bitter leaf P = Pumpkin leaf, W = Waterleaf.

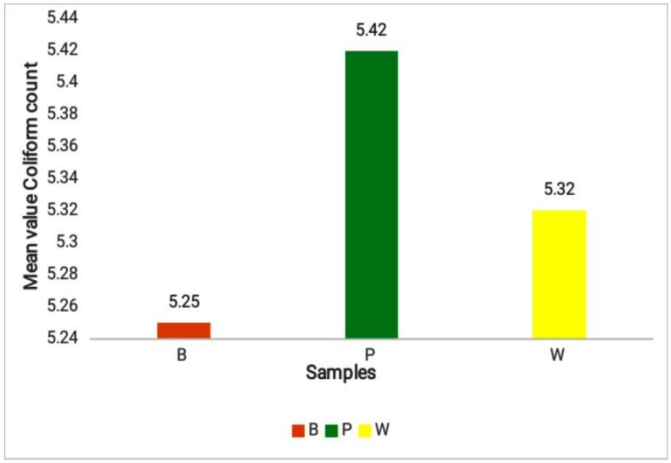

Figure-3: Total Mean of Total Coliform Count of the Different vegetables KEYS B = Bitter leaf, P = Pumpkin leaf, W = Waterleaf.

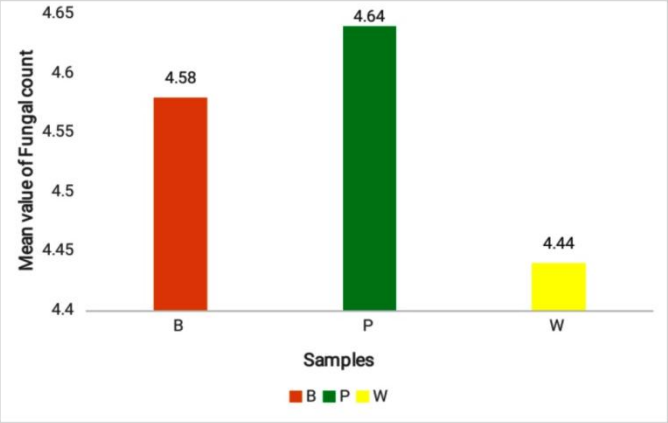

Figure-4: Total Mean of Total Fungal Count of the Different vegetable KEYS B = Bitter leaf, P = Pumpkin leaf, W = Waterleaf.

Table-2: Proximate Composition of Vegetables Samples.

Parameters	Bitter Leaf	Pumpkin Leaf	Water leaf
Ash (%)	4.35	9.32	7.81
Moisture Content (%)	3.97	8.92	12.7
Crude Protein (%)	41.99	21.95	21.47
Crude Fat (%)	6.49	6.52	5.41
Crude Fibre (%)	5.62	14.3	12.93
Carbohydrate	37.58	38.99	39.68

Discussion: Microorganisms found in various fruits and vegetables differs mostly as a result of indigenous organisms found in the soil, introduction of non-indigenous organisms through animal manure, sewage, or polluted irrigation water, transportation, and handling by the retailers[12,13].

From this study, it was observed that different vegetables had a wide range of organisms in which most of them are inherently from the soil or normal flora on the leaves.

The total heterotrophic bacteria counts gotten from the leaf samples ranged from 5.45×10^6 to 2.57×10^7CFU/g and the total coliform count ranged from 1.05×10^5 to 2.90×10^5. These values are higher than the WHO directives on microbial limits. The finding of this study is higher than the total heterotrophic bacterial count obtained by Wilson I.G.[14]. Some of the microbial counts obtained from this study conformed to the microbiological limits showing partial negligence on the part of national agencies concerned with food inspection in the country. The total Staphylococcus count gotten from this study ranged from 9.0×10^4 to 3.1×10^5CFU/g which is higher than the standard set by Food and Drug Agency (FDA) limit for Staphylococcus in food which is $<10^5$ CFU/g. Fresh vegetables are damaged as a result of physiological breakdown due to physical damage or penetration by microbes[15]. For this study, all the vegetable samples purchased from the Choba market virtually harbored one organism or the other.

The prevalence of the Enterobacteriaceae were the greatest in the three leafy vegetables their percentage occurrence are as follows; Escherichia coli 25%, Salmonella sp. 15.39%, Staphylococcus sp. 21.15%, Bacillus sp. 11.54%, Klebsiella sp. 7.69%, Shigella sp. 7.69%, Micrococcus sp. 5.77%, and Enterobacter sp. 5.77%. These organisms isolated above correlates with the work of Rajaranshi, A.[16] and Anastasiades A. et al[17]. Isolation of Staphylococcus sp. from these vegetable samples are mainly from human origin, intrinsic factors or from the environment. Staphylococcus aureus has been reported as the most prominent aetiology of pyogenic infections. Staphylococcus epidermidis is a normal flora that is found on the skin. It causes pathogenic infections in humans such as endocarditis[18]. Isolation of Salmonella sp. and Shigella sp. from vegetables are mainly because of poor personal hygiene of workers, vendors, and consumers in market places have helped in the spread of these pathogenic organisms. Washing or sprinkling vegetables with contaminated water has rendered most green leafy vegetables as a potential source of many Salmonella outbreaks. Bacillus sp. isolated from most vegetables is normal flora found in the soil. It has been reported to be pathogenic especially if found in food at a microbial load of 10^5-10^7 CFU/g[18].

The soil is the major reservoir of most microorganisms isolated from vegetables. From this study, pumpkin has the lowest microbial count of $5.45\text{x}10^6$ CFU/g, followed by bitter leaf 9.0 x 10^6 CFU/g and waterleaf has the highest microbial count of $2.57\text{x}10^7$CFU/g.

The fungal count of these vegetable samples ranged from 1.5x 10^4 to $7.1\text{x}10^4$ CFU/g. Waterleaf has the lowest fungal count of $1.5\text{x}10^4$ CFU/g, followed by bitter leaf with a fungal count of $2.65\text{x}10^4$ CFU/g, and pumpkin leaf has the highest fungal count of $7.1\text{x}10^4$ CFU/g. Fungal contamination of most vegetables is a result of the deposition of fungal spores in the air on exposed vegetables sold at the market. Airborne fungal spores are released into the atmosphere by explosive discharge mechanisms that require high humidity and moisture. These spores are carried by wind and are deposited on exposed food items at the market[19,20]. The percentage occurrence is as follows:

Aspergillus sp. 40%, Candida sp. 20%, Penicillium sp. 20%, Fusarium sp. 6.67%, and Geotrichum sp. 13.33%. The fungal isolates from this study are in agreement with the report of Nagarajan, S. and singh, D.V.[19]. The result gotten from the proximate analysis of bitter leaf is as follows; Ash (4.35%), Moisture content (3.97%) crude protein (41.99%), crude fat (6.49%), crude fiber (5.62%), and carbohydrate content

(37.58%). While that of pumpkin leaf is as follows Ash (9.32%), Moisture content (8.92%), crude protein (21.95%), crude fat (6.52%), crude fiber (14.30%) and carbohydrate content (38.99%). Waterleaf had, Ash (7.81%), moisture content.

(12.70%), crude protein (21.47%), crude fat (5.41%), crude fiber (12.93%), and carbohydrate content (39.68%). From the proximate composition of the three vegetable samples, it was observed that bitter leaf has the highest protein content, followed by pumpkin leaf, and the least was waterleaf. While for moisture content the reverse was the case. Waterleaf has the highest, followed by pumpkin leaf and bitter leaf has the least. The results of the proximate analysis of the three vegetables are in contrast to the report of the study conducted by Sodamade[21,22]. The results gotten from this study indicate that poor hygienic conditions of vegetables sold at Choba market and the consumers are at risk of food-borne infections. Though vegetables are properly de-contaminated at the point of preparation or cooking, it is important to enlighten both the vendors and consumers on the sanitory practices and health implications associated with consumption of contaminated vegetables.

The presence of Staphylococcus sp., Escherichia coli, and Salmonella sp. makes on these vegetable samples makes it a potential source for the spread of food borne pathogens. Presence of Escherichia coli, Salmonella sp. and Shigella sp. on these vegetable samples indicates faecal contamination of these leafy vegetables.

Conclusion

There is a need that farmers should follow good farming procedures to avoid self-contamination of these vegetables. Retailers and consumers should maintain good personal hygiene practices and vegetables should be properly cooked so as to reduce the microbial load before consumption.

Recommendation: It is necessary that the following points are adopted to avoid contamination of various vegetables that are sold in the market. i. Farmers should follow good farming procedures to avoid self-contamination of this vegetable by the use of wet animal dung and contaminated irrigation water. ii. Retailer/sellers of vegetables should maintain good personal hygiene so as to avoid faecal contamination of these fresh leafy vegetables. iii. Modified atmospheric packaging should be applicable in vegetables so as to avoid airborne fungal spores from contaminating these vegetables. iv. Vegetable should be properly cooked while preparing soups/dishes so as to reduce the microbial load naturally found on these fresh leafy vegetables before consumption.

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