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Research Article

Comparative Studies on Microbiological and Nutritional Qualities of Juice Produced From Pineapple

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Abstract

Analyses of juices (freshly prepared and packaged) produced from Cayenne and Red Spanish varieties of pineapple to determine their microbiological and nutritional qualities were carried out. Bacteria isolated were species of *Bacillus, Staphylococcus, Pseudomonas* and *Micrococcus* while fungi isolates were species of *Aspergillus, Fusarium, Mucor, Rhizopus* and *Penicillium.* The juice from the mixture of both varieties of pineapple had the highest total viable counts (TVC) of 4.6 x 10^2 cfu/ml. The red Spanish variety had 3.8 x 10^2 while juice from cayenne had 3.1 x 10^2. The highest coliform count of 5.0 x 10^1 was obtained from cayenne variety. However, no coliform was found in both red Spanish and the mixed pineapple juices. The mould and yeast counts in the samples were low (1.0-4.0 x 10^2). Analysis of nutritional qualities of the juice revealed that the discoloration in the three juice samples was faint yellow before pasteurization and later turned deeper yellow after pasteurization. It was observed that cayenne juice had higher pH, Total solid, Vitamin C and Brix contents than either red Spanish or the mixture of the juices. Similarly red Spanish juice had higher total titratable acidity and protein than cayenne and mixed fruit juices. However, the mixed fruit juice contained moderate amount of Vitamin C and Brix (sugar). The results

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obtained suggest that cayenne pineapple juice had poor microbiological quality but rich in nutrients.

Keywords: Pineapple, Comparative Studies, Red Spaanish, Cayenne

1. Introduction

A fruit can be defined as the ripened ovary of a flower with or without associated parts. Fruits can be classified into two broad categories; juicy fruits such as lemon, orange, lime, tangerine and pumelo and pulpy fruits such as mango, pineapple, avocado, pear, guava, pawpaw, sour soup and banana. Fruits are important in human diets due to their contributions of vital nutrients, most especially vitamin C, they are very low in fats and proteins but high in sugar as they contain large amount of glucose, fructose and sucrose. Other constituents such as organic acids, phenolic substances, volatile substances and minerals may be present and these play an important role in the chemical reactions which occur during processing and storage [1]. In addition, most fruits are often consumed fresh due to their cherished flavour/palatability and they contribute immensely to nutrients intake since no nutrients intake loss are recorded as a result of cooking as in other cooked staple foods. Fruit juice is the unfermented but fermentable natural juice intended for direct consumption obtained by a mechanical process from sound, mature fruits preserved by physical and/or chemical means [2]. Fruit juice may have been concentrated and later reconstituted with water suitable for the purpose of maintaining the essential composition and quality factor of the juice. The finished product from manufacturers is expected to have undergone proper production processes as well as good packaging that would guarantee consistent good quality of the product throughout its shelf-life [3]. Fruit juice deterioration which eventually lead to spoilage can be attributed to three major factor; enzymatic activities which are responsible for the breakdown of some essential components present in the fruit, affecting the flavor, consistency and causing browning, oxidative reactions which occur especially on interaction with gases such as oxygen, nitrogen and carbon dioxide leading to browning, reduced quality and reduced nutrient value and microbial activities which are due to the activities of moulds, yeasts and bacteria, their attack results in rapid deterioration and spoilage [4]. One of the many popular fruits juice products is the pineapple juice (*Ananas comosus*). It is a member of the tropical plants called the bromeliads and is the only edible species of that family; it is grown in hot regions all around the world. It is not a single fruit but a composite mass of between 100 and 200 berrylike fruit lets that formed together into one compact fruit. It contains an enzyme that is used to aid digestion and tenderize meat, which will not only be tender but will likely fall apart [5]. It is of two major varieties, there are cayenne and the red Spanish. The cayenne is widely cultivated in Togo among the West African countries. It is white fleshed with good aroma and flavor, the leave have smoother edge, plenty of both acid and sugar contents not only sweeter but also contains three times more vitamin C than the Red Spanish. It is more compact, smaller and more cylindrical while the Red Spanish is widely cultivated in Nigeria among the West African countries as well, it is deep yellow fleshed and a crown hard spiky leaves on top with tough shell, and it contains medium sugar and acidic contents (Moser,

1999). Fresh pineapple contains minerals such as Calcium, Chlorine, Potassium, Phosphorus and Sodium. One of the best known properties of pineapple is as a diuretic. This help to estimate toxins through the urine, helping patients with ailments of kidneys, bladder and prostate. Due to the fiber content of the pulp, pineapple prevents constipation and regularizes the intestinal flora. The present study was carried out to produce juice from Cayenne and Red Spanish varieties of pineapple fruits and to compare the microbiological and nutritional qualities of the juice produced from the two varieties of pineapple as well as to isolate and identify microorganisms present in the juices.

2. Materials and Methods

2.1. Collection of Samples

Mature, ripe healthy pineapple fruits (plate 1 and 2) were obtained from Ipodo Market in Ikeja Lagos and transported to the laboratory for extraction of the juice.

2.2. Media preparation

Commercially available media- plate count Agar, MacConkey Agar, Malt Extract Agar and Potato Dextrose Agar were used. These media were in commercial dehydrated from and were thus prepared according to the manufacturer's instructions. The resulting solution was sterilized plates. The plates were tested for sterility by incubating the freshly prepared media at 37^{o}C for 24 hours before use.

2.3. Production of pineapple juice

The pineapple were washed with a little amount of detergent and rinsed repeatedly with clean water to remove any trace of the detergent. The fruit was peeled and unwanted specks removed. It was then cut into smaller pieces with a sterile knife ready for extraction.

2.4. Extraction of juice from pineapple

The pieces of pineapple were put into sterile extractor (Kenwood, JE 500) and the extracted juice was filtered by passing through sterile muslin cloth into three sterile conical flasks.

2.5. Pasteurization of the juice

The juice was subjected to heat treatment by pasteurization at 80^{o}C for 15 minutes in a hot water bath (Grant SUB28). On cooling, it was poured aseptically into sterile bottles covered and stored at 4oC (refrigerator) for further use. Figure 1 shows the production steps.

2.7. Enumeration of microorganisms in fruit juice

This was done using the pour plate method. 1ml of the sample was aseptically introduced into sterile test tubes containing 9ml sterile distilled water to give a 1 in 10 dilution which was further diluted to 10:10 from an appropriate agar (about 45^{o}C) was poured into them, mixed and allowed to set.

2.8. Selection of pineapple fruits

Washing
↓
Sorting
↓
Peeling and cutting
↓
Juice extraction
↓
Mixing
↓
Filtering
↓
Filling
↓
Sealing or corking
↓
Pasteurization
↓
Cooling
↓
Storing

Production of pineapple juice

Source (Olusuyi, 2003).

2.9. Identification of bacteria

Following repeated subculturing, pure cultures of the different isolates were obtained, characterized and identified using biochemical tests. The isolates were identified by comparing their characteristics with those of known taxa using the schemes of Cowan (1974).

2.10. Identification of fungi

The mould and yeast isolates were stained using lactophenol cotton blue and examined under the microscope. The isolates were identified using the schemes of Cowan (1974).

2.11. Physicochemical Analysis of Fruit Juices pH

The pH was determined by weighing 30mls of each samples inde a beaker and placing it under a pH meter after it has been calibrated using phosphate buffer of pH 4.0 and bringing it to neutral at 7.0 after which the start button was pressed to determine each pH [6].

2.12. Titratable acidity

The titrated acidity was determined by measuring 25ml of each sample into a clean conical flask and titrated to a pink end point against 0.1 N sodium hydroxide (NaOH) using 2-3 drops phenolphthalein indicator. The total titrable acidity expressed as citric acid was calculated as;

Volume of 0.1 NaOH x Normality x 9

Volume of sample

Volume of 0.1 NaOH = Initial – Final

2.13. Total solid

The total solid content was determined using a hot plate with magnetic stirrer. 1 g of each samples were added into weighed crucibles and 1 ml of distilled water was added. The mixture was placed on the hot plate to dry and transferred into the oven for 30 minutes. It was thereafter kept in the desiccator to cool, weighed on the weighing meter after which readings were taken.

Final weight – weight of crucible

2.14. Quantity of sample

2.14.1. Vitamin C

The vitamin C (Ascorbic acid) content was determined by measuring 10ml of each sample into a conical flask to which 2 ml of glacial acetic acid was added. It was titrated with a solution of 2,4 dichlorophenol indophenols in the burette until permanent faint colour (which persisted for at least 15 seconds) was obtained. The titration was repeated with 5ml distilled water (blank) and 5 ml of standard ascorbic acid solution (standard). The vitamin C content of the sample was thus calculated;

T – B1 x n dilution

St – B1

Where;

T = Value of sample

St = standard solution of ascorbic acid

B1 = Blank

N = number of dilutions

2.14.2. Protein content

The protein content was determined by measuring 15 ml of concentrated H2SO4 into weighed sample. The sample was digested for 1:30 minutes with a pair blank left unfilled. 25 – 28 ml of boric acid was poured into a receiver conical flask placed underneath the distiller and filled with 10 moles of Sodium hydroxide (NaOH) and 8 ml of NaOH was dispensed into the digested bottles.

2.14.3. Brix contents

The brix (sugar) content was determined using a refractometer by adding 3 drops of distilled water and zero it. 3 drops of samples were added and readings were taken.

2.15. Sensory evaluation of fruit juice

The sensory evaluation was done by people that were relatively experienced in descriptive sensory analyses and have received no further training to know the juice that had a better flavor. The juice was tested for characteristic flavor (sweet, sweeter, sweetest), aroma, colour and general acceptability – cloudiness. foaming, swelling of container and signs of fermentation [7].

3. Results

3.1. Microbiological qualities of fruit juice

This research showed the microbial count of red Spanish juice was 3.8×10^2 cfu/ml with yeast count of 1.0×10^1, cayenne, 3.1×10^2 with yeast count of 1.0×10^2 and coliform count of 5.0×10^1. The microbial count of the mixed juice was 4.6×10^2 and yeast count of 4.0×10^1 Table 1.

3.2. Nutritional qualities of fruit juice

The pH of the three pineapple juice samples are in the table 3. From the results the pH of the red Spanish juice was the lowest, followed by that of the mixture and was highest in the cayenne juice. The discoloration was deeper in the red Spanish juice, followed by the mixture juice and lightly deep in the cayenne juice. While the discoloration was done by using the pantone colour guide, the red Spanish sample has pantone 127U while the cayenne sample has pantone 113U and the mixture sample has pantone 120U. This shows that the higher the number the deeper the colour and vice versa. The results of the titration acidity, total solids, vitamin C, protein and the brix (sugar) contents are in Table 4.

Table 1. Microbiological Qualities of the Fruit Juice

Samples	Total Viable Counts (cfu/ml)	Coliforms (cfu/ml)	Yeasts (cfu/ml)	Moulds (cfu/ml)
Red Spanish (RS)	3.8×10^2	0	1.0×10^1	0

Cayenne (CY)	3.1×10^2	5.0×10^1	1.0×10^1	2.0×10^1
RS + CY	4.6×10^2	0	4.0 x 10	0
Control (Commercial)	5.1×10^2	1.0×10^1	3.0 x10	0

Table 2. Characteristics and Identification of fungal isolates

Microscopic appearance	Microscopic appearance	possible organisms
Yellow colonies	Single celled conodia in chains developing at the end of the sterigma arising from the terminal bulb of the conidophores (vessels). Long conidiophores arising from septate mycelium	*Aspergillus niger*
Fluffy yellow colonies	small septate hyphae give rise to phialides that produce either single celled microconidia or large macroconidia that are sickle or boat shaped with numerous septa.	*Fusarium* sp
Grey colony with wooly texture	Distorted hyphae, ovoid spores, sporangia with columellas, Rhizoids present	*Rhizopus* sp
Cream coloured, Smooth, Moist colonies	Oval cells, reproduce by budding, hyphae absent	*Saccharomyces* sp

Table 3. pH and Discolouration of fruit juice analysis

Samples	pH	Discolouration
Red Spanish	4.24	very deep yellow
Cayenne	4.62	Intermidiate yellow
Mixture (RS + CY)	4.52	Deep yellow
Commercial	4.50	Very deep yellow

Table 4. Nutritional Qualities of Fruit Juices

Samples	Titratable acidity	Total solids	Vitamin C	Protein (%)	Brix (sugar)
Red Spanish	0.55	13	19	0.3996	12.8
Cayenne	0.45	15	25	0.3759	16.3
Mixture (RS + CY)	0.43	12	24	0.2005	14.1
Commercial	0.47	13	22	0.355	14.7

Table 5. Sensory evaluation of three juice samples by 10 panelists using 10 Heidonic scales

Samples	Cayenne	Red Spanish	Mixed Juice	P-value
Taste	8.80 ± 0.422	8.00 ± 0.817	6.20 ± 0.919	0.000
Colour	8.10 ± 0.568	8.60 ± 0.699	6.40 ± 1.075	0.000
Flavour	8.50 ± 0.527	7.80 ± 0.919	6.20 ± 1.033	0.000
		General		
Acceptability	8.80 ± 0.422	8.30 ± 0.675	6.20 ± 1.350	0.000

4. Discussion

The total aerobic counts of all juice samples increases, most importantly freshly made juice, during the period of storage. The same applied to proliferation of coliforms count of freshly made juice. Microorganisms isolated from this juice samples include; *Bacillus* sp, *Staphylococcus aureus* and *Pseudomonas* sp. The frequency of occurrence of *Bacillus* sp, was the highest and this agrees with the report of Splittloecter and Churey [8] that *Bacillus* is a major spoilage organism in juices. The presence of *Staphylococcus* sp in the juice is attributed to its wide spread in the environment. The primary habitat of these organisms are the interior body and skin of man and animal from where this microbes are transferred to the fruits and subsequent transfer to the fruit juice during production. *Pseudomonas* sp are commonly found on the fruit surfaces which can end up in the juice during production. They are able to grow on a wide variety of organic substrate and are regular component of food spoilage [9]. The mould and yeast isolated include; *Mucor, Aspergillus flavus* and *Saccharomyces* sp. This is in agreement with the Splittloecter and Churey, [8] who implicated fungi as contaminants of fresh fruits especially in the presence of injuries. Water and environment may play a major role in the fungi contamination of pineapple especially during washing of fruits. The presences of *Saccharomyces* sp. is expected due to its preference for sugar and low pH, which highly favor yeast proliferation [9]. The accumulation of carbon dioxide was as result of fermentation of sugar by the yeast to alcohol. Generally according to national agency for food and drug administration and control

(NAFDAC), the package juice remains drinkable even after four to five weeks which strongly oppose the freshly locally made juices. The pH value pure undiluted pineapple juice is estimated to be between 3.7 and 4.5. The slight increase in pH values of the freshly made juice with or without preservatives may be as a result of dilution with sterile, distilled water making it tend a result of the undissociated molecules which do not free the acidic portion and can therefore not have a lower hydrogen ion concentration (pH). The reduction in pH on storage is expected because juice spoilage is mainly by yeast fermentation which is characterized by higher tendency towards acidity. This agrees with the report of Frazier and Westhoff [10]. The acidity tendency of the pH is equally evident in an increasing titratable acidity value with time [11]. The total titratable acidity which is expressed as citric acid was much higher in the package juice and this correlates with the pH values as well. The higher acidity could be as a result of added organic acids such as citric, malic and sometimes ascorbic acid as acidifying agents and flavoring agents by some commercial producers [3]. For all the freshly prepared juices, the total solids value was considerably higher than the packaged juice and this may be attributed to the fine filtration methods used commercially as opposed to filtration with muslin cloth used in the present study. This agrees with the report of Densupsoontorn *et al.,* [2] that most differences in fine filtration and centrifugation means that most of the components that make up total solids in the packaged juices are most likely desired additives.

5. Conclusion

The freshly made juice from cayenne had a better nutritional quality than the juice from the red Spanish and even juice from mixture of both varieties of pineapple. With such high nutritional contents however, pineapple juices provide a good alternative to increase pineapple consumption thereby reducing wastage of the fruits. However the microbial count of cayenne had the highest which could probably be due to other environmental factors, thus pineapple juice contributes to a healthy diet. Freshly prepared juice from the combination of both cayenne and red Spanish pineapple varieties should consumed because of high nutritive value. Properly quality control measures should be used in the production (small or large scale) of pineapple juice.

References

[1] Harker, R.F., Lau, K. and Gunson, A.F. (2003). Juiciness of fresh fruit; a time intensity study. *Postharvest Biology and Technology 29(1); 55-60.*

[2] Densupsoontorn , N., Jirapinyo, P., Thomonsiri, N., Wong, R., Rhosuya, P., Tritipat, A., Pattrarat, S., Pidatcha, P., and Suwannthol, L. (2002). Comparison of the Nutrient Content of fresh fruit juices vs commercial fruit juices. *Journal of the Medical Association of Thailand* 2; 732-738.

[3] Oluseyi, A.M. (2003). Soft drinks and fruit juices. Institute of public analysts of Nigeria Newletter 4(16); 8-11.

[4] Felicia, O.K. (1995). Spoilage in fruit, Vegetable, root and tuber crops. *Nigerian FoodJournal 3 (1);* 10 7-115.

[5] Cheesbrough, M. (2000). District Laboratory Practice in Tropical Countries, Part2. Cambridge University Press. Pp 52-70.

[6] Usoro, E.U., Suyamsothy, E., Sanni, G.A. (1982). Manual of chemical method of food analysis. Food and drugs administration, federal ministry of health oshodi, lagos publication pp. 1-2, 82-84.

[7] Ihekoronye A.I. and Ngoddy, P.O. (1985). *Intergrated Food Science and Technology for the Tropics*. Macmillan Publishers Limited, London pp. 167, 171-178.

[8] Splittlocter, D.F., Lee, C.Y. and Churey, J.J. (1998). Control of *Alicyclobacillus* in the juice Industry. *Dairy Food and Environmental Sanitation* 18;585-587.

[9] Adams, M.R and Moss, M.O. (1995). *Food Microbiology*. Royal Society of Chemistry, Cambridge pp. 56 – 89, 130-131.

[10] Frazier, W.C and Westhoff, D.C. (1988). *Food Microbiology*. 4th Edition, McGraw Hill, New Delhi pp. 83-98, 196-215.

[11] Feller, P.J. (1991). The relationship between the radio of degress brix to percent acid and sensory flavor grapefruit juice. *Journal of food technology 45 (7); 68-74.*