EFFECT OF PASTEURIZATION ON THE MICROBIAL, PHYSICOCHEMICAL AND SENSORY PROPERTIES OF WATERMELON JUICE

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Purpose:The effect of pasteurization on microbial, physicochemical and sensory properties of watermelon juice were evaluated from studies conducted on three samples of watermelon juice. Method: Samples evaluated are fresh watermelon juice (FWJ), watermelon juice pasteurized by high temperature short time (PHS) and watermelon juice pasteurized at low temperature long time (PLL). Results : Microbial analysis results from the investigation showed that highest total aserobic bacteria count of 9.8 x 10^6 cfu/ml yeast and mold 15.0x 10^5 cfu/ml *Salmonella* 2.0 x10^5 cfu/ml, *Staphylococcus* 9.1 x10^5 total coliform count of 29. 0x10^6 cfu/ml were recorded in FWJ. The bacteria isolated were *Bacillus spp*, *Staphylococcus aureus* ,*Klebsiella spp and Pseudomonas spp* while the mold isolates were *Aspergillus niger and Rhizopus nigricans.* The yeast isolated were *Saccharomyces spp*, *Candida* and *Zygosaccharomyces spp*. The physicochemical analysis shows that vitamin C content was higher in the fresh watermelon juice than the pasteurized watermelon juice while the pH and the tritable acidity are not affected. The sensory evaluation shows that fresh watermelon FWJ had the highest colour 7.20 and vitamin C 6.95mg/100g. However, PHS was rated high in flavour 87 and the overall acceptability tests revealed that sample PHS was preferred. Conclusion: Studies shows that high temperature short time is an effective method of inactivating microorganism and extending the shelf-life of products.

Keywords; pasteurization, watermelon, juice, microbial , sensory

Introduction

Watermelon fruit juice is the liquid extract of watermelon fruit which are filtered to remove pulp or fiber; it is a delicacy in some countries like mexico, some of which even comes as a perfect blend with other additive fruit juice (Alam *et al*.,2013). Watermelon is a vine – like flowering plant originally from southern Africa. It belongs to the family of Cucurbitacea and the genus *Citrullus* (Inuwa, 2011). As a member of the Cucurbitacea family, it is related to the cantaloupe, squash, cucumber and gourd that grow as vines in the ground (Abdul wahal, 2011). The entire content of watermelon contains 96% water that is very sweet and refreshingly tasty

which serves the purpose of thirst quenching It contaain 7.18% carbohydrates, 0.62%protein ,0.49%ash,0.098%crude fibre and 0.43%fat(Inuwa,2011). Watermelon is an excellent source of immue-supportive vitamin C with a good amount of vitamin B6 (Pyridoxine), thiamine (Vitamin B). It is also a good source of phenolic antioxidants and carotenoid lycopene. It also contains cucurbitacin E, a triterpene anti-inflammatory phytonutrient and usual amounts of the amino acid citrulline (Dimitrovski *et al.*, 2010).

Watermelon has a lot of water which is an ideal medium for contamination by microbes and it can get contaminated during cutting and packaging with harmful bacteria and water used in fields. The juice is pasteurized prior to storage and consumption. Juice is typically pasteurized by high temperature short time (HTST) pasteurization in the fruit juice industry which uses plate heat exchanges to heat the sample quickly at 78^{o}c (Alam *et al.*, 2013).

Pasteurizing watermelon juice is necessary to preserved the juice against micro-organisms as pasteurization is the heat treatment process that destroy pathogenic micro organisms in certain foods and beverages which help to prolongs the shelf life (Cortés *et al*., 2008). It is a mild heat treatment that takes place for 30 minutes at 70 to 100% relative humidity. Spore forming bacteria that are more heat resistant require heating to 190^{o}F for several minutes but mold spores are destroyed at about 175^{o}F for 5 to 10 minutes. Yeast and acid tolerant bacteria can be killed by holding pasteurization for 15 to 20 minutes at 155^{o}F or higher. Low temperature long time (LTLT) and high temperature short time (HTST) are the most commonly used techniques for juice pasteurization. The objective of this study was to evaluate the effect of pasteurization on the microbial, physicochemical and sensory properties of water melon juice .

2. Materials and Methods

2.1 Collection and Processing of Fruits; Mature, ripe healthy water melon were bought from different sales point in Sayedero Markets, Ilaro, Ogun State, Nigeria.

2.2 Production of watermelon juice The watermelon fruit was washed with distilled water after which it was washed with 5% hypochloride solution and immediately rinsed again with distilled water. The whole fruits was cut longitudinally using a sterile stainless steel knife to avoid contamination due to corrosion and edible portion was removed, cut into small pieces,

transferred into a sterilized blender (binatone model) and blended until sufficient juice was produced. The entire slurry was transferred into a clear liquid using muslin cloth and the juice were filled in air – tight Bottle.

2.3 Sample Treatments (Pasteurization) Two bottles of watermelon juice was subjected to heat processes at low temperature long time of 63^{o}C for 30 minutes and high temperature short time of 72^{o}C for 10 minutes respectively according to (Cortés *et al*, 2008) Watermelon juice was thermally processed in a tabular stainless steel heat exchange coil 2.2mm of internal diameter plasterer and one bottle of watermelon juice sample was untreated. The three bottles were cooled and refrigerated 4^{0}C for shelflife.

2.4 Microbial Analysis Microbial isolation and identification was by standard microbiological techniques examine by day 7. Aliquot 0.1 mL of appropriate dilutions was spread inoculated in duplicate plates of Nutrient agar , MacConkey Agar, and Potato Dextrose Agar. The inocula were spread with sterile spreader to ensure even distributions before incubating the plates. Nutrient Agar was incubated at 30 for 24 – 48h. *Staphylococus ,Salmonella* and Coliform media were incubated for 35^{0}c ±2^{0}C, while Fungi (plates) were incubated at 25 ± 2°C for 3 days. Colonies were enumerated at the end of incubation period using digital colony counter (Gallenkamp, England). The isolates were characterized on the bases of colonial morphology, microscopic and biochemical characteristic (Lynne,2003)

2.5 Physio-Chemical Qualities

2.5.1 Titratable Acidity Determination :10ml of the watermelon juice sample was pipetted into 100ml measuring cylinder and it was make up to 100ml with water. 10ml of each of the sample was pipetted into different conical flask and 2drops of phenolphthalein indicator was added into the solution and was titrated with 0.1mol NaOH until a faint pink colour appear as end point. The tritration was repeated for each sample. Titration acidity was calculated as percent citric acid (AOAC, 2005).

2.5.2 Total Sugar Determination The concentration of the soluble sugar was determined using a hand held refractometer (Bellingham and stanly, model A85171). Few drops of the juice were mounted on top of the refractometer and reading were taken.

2.5.3 Total Solids Determination: This was determine through the use of Abbe 60 refractometer and was corrected to 60^0 accordingly before used.

2.5.4 P^H Measurement :The PH of the sample was determined according to the method of (AOAC 1990). 100ml of the sample was measured into 250ml beaker already cleaned and labeled. The P^H meter was first calibrated with distilled waster to 7.00 before it was immensed into each beaker containing different samples. The reading were noted and recorded accordingly.

2.5.5 Specific Gravity Determination: An empty bottle was initially weighed and recorded as (Ag) and water was placed in the bottle and recorded as (Bg). The weight of the sample and the gravity bottle was also determined and recorded as (Cg). Specific gravity of each sample was calculated as follows

$$\text{Specific gravity} = \frac{C - A}{B - A}$$

2.5.6 Ascorbic Acid (Vitamic C) Determination; Standard ascorbic acid solution 10mg in 50ml of water preparation of indophenols dye, 20g of the sample was weighed into 100ml of water in a volumetric flask. 25ml of 20 percent metaphosphoric acid was added to 100ml with diluted water. 10ml of the standard solution was pipette into a conical flask and 25ml of acetone and titrate against indophenols solution until pink colour persist for second. Ascorbic acid constant will be calculated as mg/100g of sample and 10ml of watermelon juice was pipette into conical flask and titrated immediately with standard solution of 2,6 dichlorophenol to a faint pink end point. The procedure was carried out in triplicate for all the samples.

The ascorbic acid content was calculated as follows:

$$\text{Ascorbic Acid (mg/100ml)} = \frac{VXT}{W} \times \frac{100}{1}$$

Where V = Titre value of sample

T = Ascorbic Acid equivalent to ldm^3 of ascorbic acid used W = Volume of sample used.

2.6 Sensory Evaluation

The method of lhekoronye and Ngoddy (1985) were used. The watermelon juice samples was evaluated for the following parameters; taste, colour, flavour and overall acceptability by a panel of fifteen judges (using a questionnaire) of regular fruit juice consumers using the Hedonic scale. The sensory scores were analysed statistically using Duncan multiple range.

Results And Discusssion

3.1 Results

Table 1: Microbial Analysis Of Watermelon Juice

Sample	Total Viable Count (cfu/ml)	Total Coliform Count (cfu/ml)	Yeast and mold (cfu/ml)	Salmonella count (cfu/ml)	Staphylococcus count (cfu/ml)
FWJ	9.8×10^6	29.0×10^6	15.0×10^5	2.0×10^5	9.1×10^5
PLL	7.2×10^5	5.0×10^5	3.2×10^5	NL	3.2×10^5
PHS	4.6×10^5	2.0×10^5	1.5×10^5	NIL	1.0×10^5

KEY: FWJ -Fresh untreated watermelon juice
PLL -Watermelon juice pasteurized at Low temperature longtime (60°C for 30 minutes)
PHS -Watermelon juice pasteurized at high temperature short time (72°C for 10 minutes
Day O .no microbes observed
Day 7. microbial analysis shown above

Table 2: Morphological Characteristics Of Suspected Organisms Isolated from Watermelon Juice

Suspected Micro-Organisms	Colour	Gram Reaction	Shape	Catalase test	Oxidase test	Coagulase	Glucose	Sucrose	Lactose
***Bacillus* spp**	Cream	+ve	Rod	-ve	+ve	-ve	+ve	+ve	+ve
***Pseudomonas* spp**	Cream	-ve	Rod	+ve	-ve	+ve	+ve	+ve	+ve
Staphylococcus spp	Orange	+ve	Cocci	+ve	-ve	+ve	+ve	+ve	+ve
***Lactobacillus* spp**	Cream	+ve	Rod	-ve	-ve	-ve	+ve	+ve	+ve

Table 3: Physico-Chemical Quality Assessment Of Watermelon Juice

Sample	pH	TTA (%)	TSS (%)	Vitamin C (mg/100g)	Specific gravity	Refractive index (%)
FWJ	4.4± 0.02	0.18± 0.01	5.50± 0.00	6.95± 0.02	0.99± 0.01	1.34± 0.03
PLL	4.10 ± 0.01	0.10± 0.03	4.50± 0.01	5.36± 0.09	1.01± 0.09	1.34± 0.01
PHS	4.00± 0.01	0.11± 0.01	4.00± 0.01	4.50± 0.09	1.00± 0.01	1.33± 0.04

Value are means of triplicate determination

Table 4: Sensory Evaluation Of Watermelon Juice

PARAMETER	**Colour**	**Flavour**	**Taste**	**Over all acceptability**
FWJ	7.20^{b}	5.93^{a}	4.27^{b}	5.13^{b}
PHS	5.66^{a}	6.87^{a}	6.13^{a}	7.73^{a}
PLL	5.73^{a}	6.20^{a}	6.73^{a}	7.20^{a}

Values with the same superscripts within a column has no significant different

4.0 Discussion

Table 1 above show the microbial analysis of watermelon juice pasteurization at different time – temperature and fresh watermelon juice at the 7day. The total aerobic bacteria count for sample FWJ, PLL and PHS are 9.8 x 10^6, 7.2 x 10^5 and 4.6 x 10^5 cfu/ml respectively. The coliform count range from 29 .0x10^6 to 2.0x10^5 cfu/ml, the yeast and mold count range from 15.0 x 10^5 to 1.5 x 10^5 cfu/ml respectively. The salmonella count for sample FWJ was 2.0 x 10^5cfu/ml and no count detected in the pasteurized samples while staphylococcus count range from 7.1 x 10^5 to 1.0 x 10^5 cfu/ml respectively. It could be deduced that the fresh watermelon sample FWJ has the highest microbial counts.

The morphology characteristic of microbial isolated in the watermelon juice are presented in table 2. The result revealed that the bacterial contaminants of watermelon juice samples were identified as species of *Bacillus, Pseudomonas, Klebsiella and Staphylococus* while the mold isolates were *Asperigillus niger, Mucor sp, Rhizopus and the yeast identified are Saccharomyces cerevisiae, Zygosaccharomyces* spp *and Candida* spp. Bacillus species were more frequently encountered, this agrees with the report of (Splltttstoesser *et al*., 1994 and Bello *et al* .,2014) that Bacillus is a major spoilage organisms in juice. The presence of *Staphylococcus aureus* in the juice could be attributed to its wide spread in the environment (Dai *et al*., 2006). The occurrence of *klebsiella sp* could have been as a result of contamination from equipment, since it is a common equipment contaminant.

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 substrates and are regular components of food spoilage (Adams and moss, 1995). The presence of coliforms in these juice could be due to the high water activity of ready-to-serve juice. Products with high water activity possess good amount of unbound water molecule that support growth and survival of micro organisms (Antony and Chandra, 1997, Ferrati *et al*, 2005).

The physicochemical quality assessment of the juice sample are shown in table 3. The study revealed that the pH of the freshly made watermelon juice and pasteurized watermelon juice range from 4.00 – 4.40. This result agrees with the result obtained by Adams (1996) who reported that pH value of most fruit juice is estimated to be within the range of 2.5 – 4.4.

The titratable acidity was higher in freshly made juice than the pasteurized juice sample. The total solids of freshly made juice, low pasteurized and high pasteurized watermelon juice are 5.50, 4.50 and 4.0 respectively. The total solid and juice content are used in characterizing the quality of juice and other beverage products. (Adubofur *et al.,* 2010). The vitamin C content in freshly made juice was higher than the vitamin C content in pasteurized juice. The vitamin C content range from 6.95 – 4.50 mg/100g, which demonstrate that vitamin C concentration of watermelon juice was affected by both time and temperature. Vitamin C is a heat labile and is quickly degraded during thermal pasteurization. The specific gravity of the juice range from 0.99 – 1.00 and the refractive index range between 1.33 – 1.34.

The results of the sensory evaluation of the fruit juice samples is showed in table 4 which indicate significant difference ($P < 0.05$) for all watermelon juice quality parameters when the sample were untreated (freshly made watermelon juice),Pasteurized juice at 72^{o}C of 10mins and pasteurized juice at 63^{o}C for 30minutes. Colour was one of the most affected parameter after pasteurization which decreased in sample PHS and PLL as 5.66 and 5.73 respectively as a result of thermal processed. The untreated juice was rated high in colour due to lycopene retention in the juice. An attractive red colour is one of the primary characteristic of watermelon juice so browning is a quality handicap (Aguilo-Aguaya *et al*., 2010).

The result revealed that sample PHS scored highest in term of overall acceptability and flavour and sample PLL was rated high in term of taste. However the untreated watermelon juice FWJ was not preferred because of flavour and taste which was been affected during storage due to microbial decomposition of the fruit juice.

Conclusion

From the study it can be concluded that high temperature short time (HTST) pasteurization is an effective method of inactivating micro organism and enzymes. Although it can cause detrimental effects on the quality of the juice. Heat treatment causes colour change, separation of particle and change in flavour or smell. It also revealed that fresh watermelon juice was more susceptible to microbial attack compared to pasteurized juice which was stable till 2 weeks.

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