

Microbiological Quality Assessment of Commercially and Laboratory Prepared Orange Juice

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Abstract

Orange juice has become one of the most widely accepted natural beverages due to its invigorating flavours, source of quick energy in the form of natural sugars and its nutrient which support the growth of acid tolerant bacteria, yeasts and moulds. Present investigation was conducted to study the microbiological examination of commercially and freshly prepared orange juices. Freshly prepared orange juices were pasteurised, carbonated, concentrated and chemically treated with a preservative (sodium metabisulphite) and stored under, freezer, refrigeration and ambient temperature for 90 days. The pH ranged from 3.0 to 4.5 for the commercially, fresh and stored samples respectively. The total bacterial load ranged from 1.6×10^5 to 3.6×10^6 and the total yeast cell counts ranged from 1.7×10^4 to 4.8×10^6 cfu/ml for the commercially, freshly and stored samples. Twenty-two microbial species including 5 bacterial isolates, 6 yeast isolates and 11 isolates of mould were isolated from the orange juices. The bacterial isolates identified from the samples were *Bacillus megaterium*, *Bacillus cereus*, *Bacillus pantothenicus*, *Bacillus aeruginosa* and *Escherichia coli*. The fungal isolates were *Saccharomyces cerevisiae*, *Saccharomyces rouxii*, *Saccharomyces telluris*, *Blastomyces* sp, *Aspergillus* sp, *Mycelia* sp, *Chrysosporium* sp, and *Trichoderma* sp. Among the bacterial isolates, *Bacillus* sp was the predominant, while the fungi *Saccharomyces* sp and *Mycelia* sp were the predominant fungi and accounted for five out of 11 isolates. The isolates of bacteria, yeasts and moulds appear to be persistent throughout the period of this study and could be used as indicators of microbial quality. A safe microbial load and reduction in contamination of orange juice /fruit juices can be achieved by combination of processing methods such as chemical preservatives with pasteurisation, concentration with carbonation under controlled microbiological environments such as freezer and refrigeration temperatures.

Keywords: Orange juices; Processing; Microbial load; Microbial quality; Microbial contamination.

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1. Introduction

Consumption of fresh juices increased drastically due to its freshness, high vitamin content and low calories [1]. It is prepared by mechanically squeezing or macerating without the application of heat or solvent [2] and may in the home from fresh fruits and vegetables using variety of hand and electric juicers [3]. One of the limiting factors that influence the fruits economic value is the relatively short shelf-life period caused by pathogens attack [4]. It is estimated that about 20-25% of the harvested fruits are destroyed by pathogens during post-harvest handling even in developed countries [4-6]. In developing countries, post-harvest losses are often more severe due to inadequate storage and transportation facilities [4]. Pathogenic organisms can enter fruits and vegetables through damaged surfaces, such as punctures, wounds, cuts and splits that occur during growing or harvesting [7].

Yeasts and moulds are favoured as spoilage agents of fruits juices compared to bacteria because of the physical and chemical properties of fruit juices [8, 9]. Some of these properties include the low pH of the fruit juices, the positive oxidation reduction potential, water activity of the fruit juices and the rich nutrient composition of the juices [8, 9]. The spoilage caused by microorganisms in juices include cloud loss, development of off-flavours, carbon (IV) oxide production and changes in colour, texture, and appearance resulting in degradation of product [10, 11]. The most commonly reported bacterial genera include *Acetobacter*, *Alicyclobacillus*, *Bacillus gluconobacter*, *Lactobacillus*, *Leuconostoc*, *Zymomonas*, and *Zymobacter*. Among yeasts, *Pichia*, *Candida*, *Saccharomyces* and *Rhodotorula* are commonly encountered genera responsible for spoilage of juices [12]. Certain common moulds such as *Penicillium* sp, *Aspergillus* sp, *Eurotium* sp, *Alternaria*, *Cladosporium*, *Paecilomyces*, *Botrytis*, *Colletotricum* and *Curvularia* have been reported in spoilage of a variety of fruit juices [10, 13].

Quality in the context of fruit juices (orange juice) is the sum total of all those attributes which come to make fruit juices acceptable, desirable and nutritionally valuable as human food [14-16]. Microbiological quality of juices

or drinks is ascertained in order to ensure the safety of the consumer. Traditionally, the detection and enumeration of indicator organisms rather than of pathogens have been used. The coliform has the principal indicator of the suitability of a particular drink for consumption [17]. Indicator organisms in food serve as a tool to evaluate the microbial quality of the product. The aerobic plate count also gives a useful measure of the quality of the product and allows prediction of the shelf-life and also the identification for potential hazards by the use of suitable indicators or by direct detection of pathogens [14].

In developing nations like Nigeria, fruit juices account for more than 90% of the total fruit production [3]. It has been known that fruit constitute commercially and nutritionally important indispensable food commodity [4]. Fruits play a vital role in human nutrition by supplying the necessary growth factors such as vitamins and essential minerals in human daily diet and that can help to keep a good and normal health [4].

This present study therefore investigates the microbiological safety of commercially and laboratory processed orange juices. This would provide a background microbiological data for development of methods that would effectively reduce the microbial load and contamination of fruit juices including those considered to constitute spoilage threat and potential health hazard to consumers.

2. Materials and Methods

2.1. Sample Collection and Preparation of Orange Juices

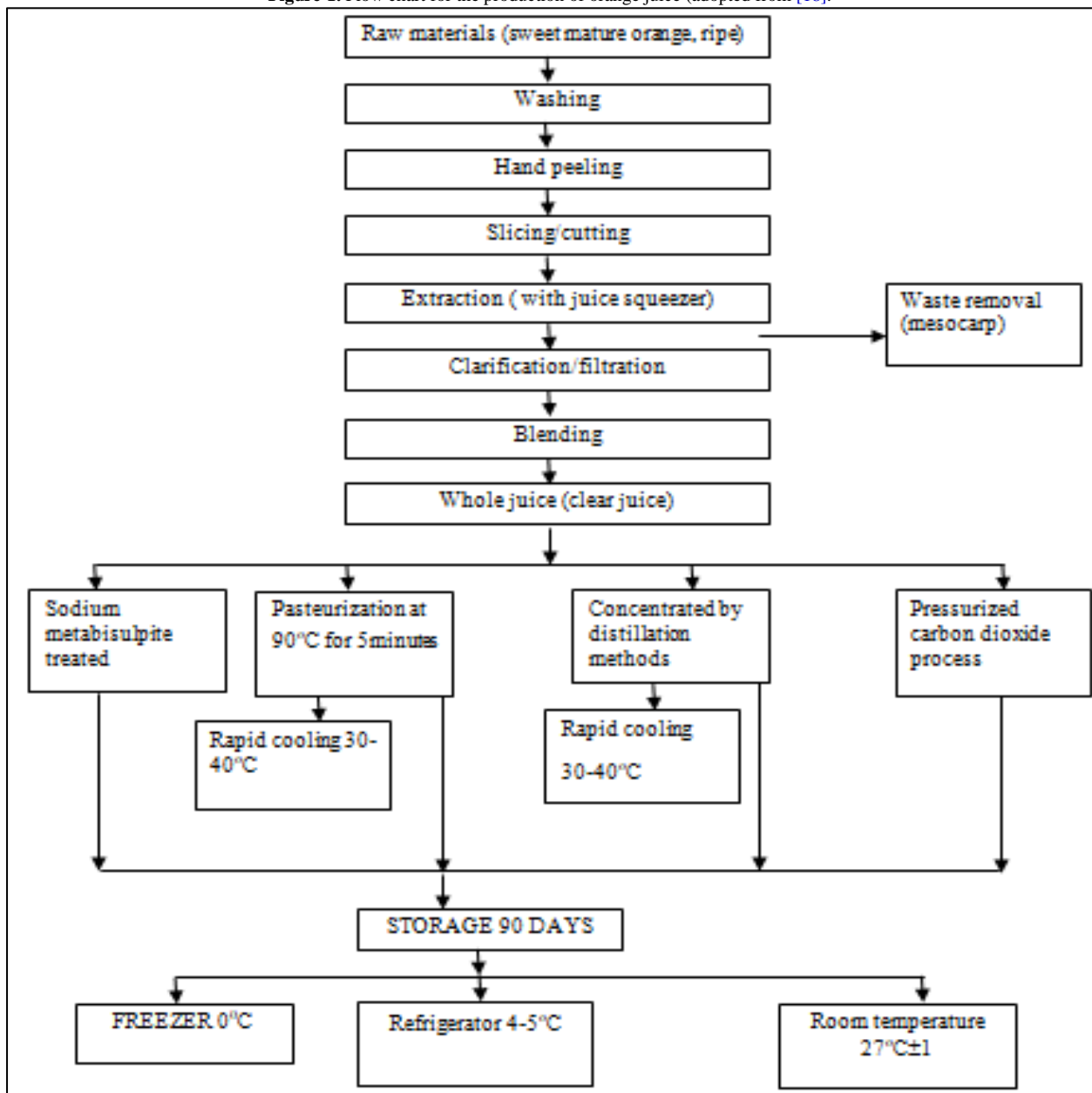
A total of 14 orange juice preparations comprising of 12 treated samples, one commercially processed juice were used for the study as indicated in (Table 1). Mature ripe sweet oranges (*Citrus sinensis*) and commercially processed orange juice were purchased from the market in Kaduna State, Nigeria. Using the processing method of AKpapunam, *et al.* [18], with some modifications, six hundred medium sized intact sweet oranges (*Citrus sinensis*) were selected, washed, peeled and sliced into halves. The cut oranges (mesocarp) were pressed with a hand juicer squeezer to extract the juice. The juice and pulp obtained was homogenized in a sterile hand monilex blender. The homogenate was filtered with sterile cheese cloth to obtain a whole juice (clear juice) (Figure 1). This was divided into five batches of 400 millilitres each and given different treatments as shown in (Table 1). Each batch of the four hundred millilitres (400ml) of the extracted juice was pasteurised (PASD) at 90°C for 5 minutes in a water bath, carbonated juice (CAB) was treated with carbon (iv) oxide gas at a concentration of 1.0kg/100ml of orange juice with a pressure of about three atmosphere at 10°C. The concentrated juice (COND) was concentrated by heating to boiling with distillation apparatus and chemically (CHM) treated with a preservative (sodium metabisulphite 0.035gm/100ml of juice). The freshly prepared laboratory (FRH) and commercially processed (COS) orange juices were used as control samples. The pasteurised and the concentrated samples were allowed to cool to about 30-40°C. The treated samples of CHM, CAB, COND, and PASD were stored for 90 days under freezer, refrigerator and ambient temperature (Figure 1).

Table-1. General Characteristics of the Fruit Juices used for the Assay

S/no	Sample code	Type of Treatments
1	*FRH	Fresh laboratory (lab) produced orange juice
2	*COS	Commercially processed orange juice
3	CHM ₁	Chemically treated lab produced orange juice stored in the freezer
4	CHM ₂	Chemically treated lab produced orange juice stored in the refrigerator
5	CHM ₃	Chemically treated lab produced orange juice stored in an ambient temperature
6	CAB ₁	Carbonated lab produced orange juice stored in the freezer
7	CAB ₂	Carbonated lab produced orange juice stored in the refrigerator
8	CAB ₃	Carbonated lab produced orange juice stored in an ambient temperature
9	COND ₁	Concentrated lab produced orange juice stored in the freezer
10	COND ₂	Concentrated lab produced orange juice stored in the refrigerator
11	COND ₃	Concentrated lab produced orange juice stored in an ambient temperature
12	PASD ₁	Pasteurised lab produced orange juice stored in the freezer
13	PASD ₂	Pasteurised lab produced orange juice stored in the refrigerator
14	PASD ₃	Pasteurised lab produced orange juice stored in an ambient temperature

**Control samples.

Figure-1. Flow chart for the production of orange juice (adopted from [18]).



Measurement of pH: The pH of juice samples were measured using a pH meter.

Microbiological analysis: This was determined according to the method of Lateef, *et al.* [19], for the enumeration of microorganisms from fresh, pasteurised, carbonated, chemically treated and the commercially processed orange juices. Each of the various samples of orange juice were analysed by serial dilution agar plate technique. An aliquot of ten millilitres (10ml) of each of the orange juice samples were mixed with ninety millilitres (90ml) of sterile peptone water and homogenised by manual shaking. Each sample was serially diluted (10 fold dilution of 10^{-1} , 10^{-2} , 10^{-3} up to 10^{-10} dilutions) in sterile peptone water. 0.1ml of each of the diluent was plated on the various agar media in duplicate for enumeration for bacteria and fungi. Plates of PCA were incubated at 37°C for 24-48h for enumeration of total viable counts for bacteria, while enumeration of yeasts and moulds were done after three to four days. All enumerations were expressed as colony forming units per millilitre (cfu/ml).

Identification of Bacteria: The bacterial isolates were identified following standard microbiological procedures as described by Cheesbrough [20] and Kampfer, *et al.* [21]. The purified bacterial isolates was observed under microscope by Gram stain method and further various biochemical tests were performed for identification of bacteria such as catalase test, oxidase test, starch hydrolysis test, IMViC test and sugar fermentation test [21].

Identification of Yeasts and Moulds: The methods adopted for identification of yeast include morphological characteristics, fermentation of sugars and urea hydrolysis as described in “Fungi and food spoilage” [22]. Moulds were identified on the basis of morphological and cultural characteristics such as colour of colony, surface, appearance, presence and absence of cell walls, asexual and sexual reproductive structures. Further identification of moulds was carried out according to the methods described in “fungi and food spoilage” [22]. Moulds were cultured on Czapek Yeast Extract agar (pH6.7) at 25°C

3. Results

The number of bacteria and yeast cells counts from different juices stored under different storage conditions: freezer, refrigeration and ambient temperatures are shown in (Table 2). It shows that the total aerobic plate counts (TAPC) per ml for all the samples for bacteria ranged from 1.50×10^5 to 4.1×10^6 cfu/ml. The bacterial counts for the carbonated and pasteurised sample stored in the freezer had the highest counts of 8.1×10^5 to 9×10^5 cfu/ml. The chemically treated, carbonated, concentrated and pasteurised samples stored under refrigerator had counts of 3.75×10^6 cfu/ml respectively. The samples stored under the ambient temperatures had counts of 1.50×10^6 , 1.73×10^6 , 2.7×10^6 and 3.4×10^6 cfu/ml for the chemically treated, carbonated, concentrated and pasteurised samples. The fresh and commercial samples had 1.6×10^5 and 4.40×10^5 cfu/ml. Total plate count for yeast cells ranged from 1.7×10^6 to 4.75×10^6 cfu/ml. The samples stored in the freezer had the counts of 5.95×10^5 , 1.99×10^6 , 6.4×10^5 and 5.75×10^5 cfu/ml for pasteurised, carbonated, concentrated and chemically treated respectively. The refrigerated samples had the counts of 1.7×10^4 , 1.7×10^4 and 4.75×10^6 cfu/ml for concentrated, carbonated, chemically treated and pasteurised respectively, while the counts for the samples stored under ambient temperatures had counts of 8.0×10^5 , 9.2×10^4 , 2.5×10^6 and 3.4×10^6 cfu/ml for carbonated, chemically treated, pasteurised and concentrated respectively. The fresh and commercial samples had counts of 8.5×10^4 and 5.5×10^4 cfu/ml respectively.

The results of the bacterial and fungal isolates based on gram reaction, cultural, morphological and biochemical tests are shown in Tables 3, 4 and 5. A total of 22 microbial species including 5 bacterial isolates, 6 yeast isolates and 11 mould isolates were isolated from the juices classified by grouping them into 2 bacterial species, 4 yeast species and 7 mould species. The isolates of bacterial species had the *Bacillus* as the predominant genus and these are *Bacillus megaterium*, *Bacillus cereus*, and *Bacillus pantothenicus*. Others include *Pseudomonas aeruginosa* and *Escherichia coli* (Table 3).

Yeast isolated were *Saccharomyces cerevisiae*, *Saccharomyces telluris*, *Saccharomyces rouxii*, *Rhodotorula mucilaginosa*, *Brettanomyces anomalus* and *Candida mesenterica* (Table 4).

The species of mould isolates included *Mycelia* sp, *Helminthosporium* sp, *Blastomyces* sp, *Chrysosporium* sp, *Aspergillus* sp and *Trichoderma* sp (Table 5). The *Mycelia* sp was the predominant genus having accounted for five out of 11 isolates.

Table-2. Microbial load of various types of orange juices

S/no	Sample code	Total Aerobic Plate Count (cfu/ml) of Bacterial cells/ml	Total Plate Count of Yeast cells/ml (cfu/ml)
1	*FRH	1.6×10^5	8.4×10^4
2	*COS	4.40×10^5	5.5×10^4
3	CHM ₁	1.68×10^6	5.75×10^5
4	CHM ₂	3.75×10^6	1.7×10^6
5	CHM ₃	1.50×10^6	9.2×10^4
6	CAB ₁	8.1×10^5	1.99×10^6
7	CAB ₂	3.11×10^6	1.77×10^6
8	CAB ₃	1.73×10^6	8.0×10^5
9	COND ₁	3.75×10^6	6.4×10^5
10	COND ₂	4.1×10^6	1.7×10^4
11	COND ₃	2.7×10^6	3.4×10^6
12	PASD ₁	9.1×10^5	5.95×10^5
13	PASD ₂	3.4×10^6	4.75×10^6
14	PASD ₃	3.4×10^6	2.58×10^6

**=Control samples.

Table-3. Biochemical characteristics of isolates of bacteria from commercially and laboratory produced orange juices test results of isolates of bacteria

Isolates code	Catalase	Citrate Utilization	IMViC Test		Nitrate Reduction	Starch Hydrolysis	Hydrogen Sulphite Production	Acid Production	Inference (Organisms)
			Methyl Red	Voges Proskauer					
PASD01, CAB01, FRH01, CHM01,	+	+	+	-	-	+	-	A	<i>Bacillus megaterium</i>
CHM02, CAB02, FRH02,	+	+	-	+	+	+	-	A	<i>Bacillus cereus</i>
CHM03, PASD02, CAB03	+	-	+	-	-	+	-	A	<i>Bacillus pantothenicus</i>
CHM04, COS01, CAB03	+	-	+	-	-	-	-	A	<i>Pseudomonas aeruginosa</i>
COS02, CHM05	+	-	-	-	-	-	-	A	<i>Escherichia coli</i>

KEY: + → Positive, - → Negative, TSI → Triple Sugar Iron Agar

Table-4. Yeast Isolate Identity

Isolate code	Fermentation of Sugars							Identity of Yeast
	Glucose	Galactose	Maltose	Sucrose	Lactose	Melibiose	Raffinose	
FRHW,	+	-	+	-	±	-	-	<i>Saccharomyces cerevisiae</i>
CAB ₂ W, CHM ₂	+	-	+	+	±	-	-	<i>Saccharomyces rouxii</i>
COND ₂ W								
PASD ₁ P	+	-	-	-	±	-	-	<i>Saccharomyces telluris</i>
CHM ₁ P, FRHP								
CAB ₃ , FRHW	-	-	-	-	±	-	-	<i>Rhodotorula mucilaginosa</i>
FRHP, PASD ₁ P	+	-	-	+	±	-	-	<i>Brettanomyces anomalus</i>
CHM ₃ , COND ₁								
PASP ₃ , COND ₃	+	-	-	-	±	-	-	<i>Candida mensesteric</i>
CAB ₁ P, FRHP								

Key: + = Positive result, - =Negative result, ± Doubtful

Table-5. Microscopic and colonial morphology of mould isolates from commercially and laboratory processed orange juice

Sample Code	Isolate Code	Microscopic Morphology	Colonial Morphology	Inference
COS CAB ₂ CHM ₃ COND ₃ PASD ₁	A	Irregular groups of cells or hard masses of hyphae without spores	Colonies are widely spread, white and pink mycelium cell masses. Reverse is colourless	<i>Mycelia</i> sp
COND ₃	B	Multiseptate conidia with smooth walled conidiophores	Colony growth is rapidly spreading, slight pink and wooly with black reverse	<i>Helminthosporium</i> sp
CAB ₁	C	Spherical conidia borne on short hyphae branches	Colonies are dimorphic in shape. The entire surface is covered with a white wooly mat	<i>Blastomyces</i> sp
PASD ₁	D	Simple branched conidiophore in chains	The surface is rough. Woolly dark green. The reverse is pale to colourless	<i>Gliocladium</i> sp
PASD ₁	E	Irregular branched conidiophores. Some appear singly and some in short chains	Colony growth appear waxy to velvety and look pink in colour. The reverse is colourless	<i>Chrysosporium</i> sp
PASD ₁	F	It has swollen vesicle at the tip of the conidiophores	Colonies are fine velvety wooly or cottony aerial hyphae with abundant mycelium. Reverse is colourless. Conidia heads are blackish	<i>Aspergillus</i> sp
CAB ₃	H	Branched conidiophores some appear singly and some in groups	Colony shows white hyphal growth with compact woolly tufts looking green	<i>Trichoderma</i> sp

4. Discussion

Many microorganisms are found in fruit juices and soft drinks as environmental or raw material contaminants either during their growing in fields, orchards, vineyards or greenhouse or during harvesting, post-harvest handling and distribution [2]. But relatively few can grow within the acidic and low oxygen environment, yeast are the most significant group of microorganisms associated with spoilage of fruit juices and soft drinks [2]. Most fruits contain bacterial counts of 1×10^5 cfu/ml on their surface [7, 23-25]. Improper washing of fruits add these bacteria to juices leading to contamination [7]. The result obtained from this study showed that all the orange juices tested had a microbial count which was relatively high ranging from 1.5×10^6 to 4.1×10^6 for bacteria and 1.7×10^6 to 4.75×10^6 cfu/ml for yeast of the orange juices under different processing methods and storage conditions. The control samples freshly prepared and commercially processed also had high counts of 1.6×10^5 and 4.4×10^5 cfu/ml

respectively. The microbial counts of bacteria, yeasts and moulds in the samples exceeded the maximum recommended standards by the International Commission on Microbiological Specification of Foods International Commission on Microbiological Specifications of Food (ICMSF) [26], Food and Agricultural Organisation (FAO) [27] and National Agency for Food and Drug Administration and Control News (NAFDAC) [28] ; SON- Standard Organisation of Nigeria [29].

According to these agencies, the acceptable limit of mesophilic aerobic bacteria and fungi in dried foods, fruit juices and soft drinks should not exceed a maximum of 10^3 cfu/ml and 2×10^4 cfu/ml respectively. However, the microbial counts obtained in this study are considerably high since no microorganisms should be recovered in any food meant for human consumption [27, 30, 31]. The samples could therefore constitute a health hazard to consumers. The high microbial counts of bacteria, yeasts and moulds could be attributed to the fact that fruits mainly are contaminated with various microorganisms at the harvest time mostly from soil and dust [7]. Also, a high viable count often indicates unsatisfactory sanitation, unsuitable time or temperature conditions during production or storage [32, 33]. The generally observed high microbial counts in this study could be attributed to the influence of environmental factors on the microbial population which have been shown to play a significant role in affecting the quality of food products [30, 34-38].

The microbial population consisted of *Bacillus* sp, *Saccharomyces* sp and *Mycelia* sp as the most predominant isolates for bacterial and fungal isolates. The presence of some of these organisms are not surprising as most of them are known to thrive in medium rich in fermentable substrates such as sugars which often led to the production of acids after fermentation [39]. Growth and metabolic activities of these organisms contributed to the spoilage of the orange juice in storage. Fruit juices and concentrates are known to contain adequate nutrients which can support microbial growth [40, 41]. Use of these nutrients by the microorganisms could result in production of substances such as diacetyl and acetyl methyl carbinol [14, 40, 42]. These substances will influence the pH of the juice and cause spoilage. Reports by some workers have shown that pH could be used as indicator of spoilage of fruit juices [10, 11, 13, 43]. Moreover, studies have shown that moulds and yeasts tolerate high osmotic and low pH and grow at refrigeration temperatures and can therefore cause spoilage in processed product [44]. Reports by Parish, *et al.* [45]; Ghenghesh, *et al.* [46] and Raybaudi-Massilia, *et al.* [1] showed that bacteria species of *Pseudomonas* and *E. coli* can grow in orange juice, apple juice and pineapple squash. Food Safety and Hygiene discussed the survival of Enterohaemorrhagic *E. coli* (EHEC) and *Salmonella* species in fruit juices (pasteurised orange juice). They concluded that EHEC and *Salmonella* can survive in fruit juices and some other acid foods with a pH below 4.5 for many days especially at refrigeration temperature [47]. These reports confirms the results of this present study where *Pseudomonas* and *E. coli* was among the bacterial isolates being isolated from the orange juice samples commercially, pasteurised, chemically treated and stored in the refrigerator and at ambient temperatures respectively. Spores of *Bacillus* were isolated from various Egyptian canned juices and vegetables fruit juices and bottled drinks in a study by Essien, *et al.* [39], Abdalla, *et al.* [48], Gabriel and Abdul [49]. *Bacillus* species are spore formers whose spores could survive high temperatures of processing [39]. The thermoduric nature of the spores of these microbes ensure survival at pasteurisation temperatures [39]. These reports thus, confirms the possible isolation of the species of *Bacillus* being isolated from the orange juice sample analysed especially, the pasteurised and those stored in the freezer, refrigerator and ambient temperatures. Reports by Beech and Davenport [50] showed that species of *Bacillus* and *Pseudomonas* can survive freezing temperatures. The high acid and sugar content of fruits often permit yeasts and moulds to predominate in fruit juices [8, 51]. *Mycelia* species were predominant mould contaminants of the samples tested. The yeasts and mould species isolated from the test samples were mainly from the samples stored in the freezer and this shows that fungi are more likely to grow during slow thawing especially yeasts. This is because the process of freezing cannot destroy them [7]. This result is in agreement with the reports of previous studies [7, 52]. They reported that when ice is thawed, the surviving microorganisms though may be injured, tend to recover their viability so that when ice melts into the juices, they may be able to survive these too ([7, 52]. The detection of *Candida* sp, *Saccharomyces* sp, and *Brettanomyces* sp as yeasts contaminants of fruit juices especially orange juice agrees with the report of Covadonga, *et al.* [44], International Commission on Microbiological Specifications of Food (ICMSF) [26]; Renard, *et al.* [53], Obire, *et al.* [8]. *Saccharomyces* sp have been known to be responsible for large scale spoilage while slow spoilage has been caused by members of the genera *Brettanomyces* and *Candida* as reported by Bevilacqua, *et al.* [12], Kamal, *et al.* [13] in fruit juice and soft drinks. Statistically, there were no significant differences in numbers of bacteria and yeasts obtained in the test samples and in the control ($p \geq 0.05$).

Sources of fruit and fruit juices are commonly contaminated with yeasts and moulds often from the insect damage. Fallen fruits should thus be avoided where possible [2]. In terms of mould growth sources of contamination could be from spores of mould in the environment or from additives used since it is known that some of these additives deteriorate with time if they were not properly sealed hence the growth of moulds. This is evidently seen in the present study where the level of sodium metabisulphate of 350ppm (0.035%) source of sulphur dioxide were ineffective in inhibiting yeast and moulds growth in the orange juices in storage at pH values of 3.0-3.5 and even 4.5. Although Beuchat [54] in his investigation using several antimycotic agents found sulphur (IV) oxide to be inhibitory against *Byssoschlamys niver* ascospores amongst the antimycotic agents tested. Therefore, it would not be a good preservative for fruit juices especially orange juice and soft drinks in Nigerian markets since many species of yeasts and moulds were isolated from the orange juice samples tested in storage.

5. Conclusion and Recommendations

Nigerian processors of fruit juices employ techniques similar to the ones that was adopted in this study. These techniques are pasteurisation, carbonation, concentration and use of chemical preservatives and their products being stored under similar methods applied to this study: freezer, refrigeration and ambient temperatures. The processes were designed among other things to prolong shelf-life of the products by controlling microbial contamination and growth.

The average counts for bacterial and fungal isolates in the tested orange juice samples showed counts far above the maximum allowable limit in foods to be marketed for consumption. These high counts are suggestive of heavy contamination and indicate a public health concern.

The study also showed the presence of large number of bacterial and fungal species among which *Bacillus* sp, *Mycelia* sp, and *Saccharomyces* sp were the most predominant. *Saccharomyces* sp isolated from these orange juice samples can be screened for leavening ability. Thus, the use of good quality raw fruits is essential to the production of a high quality product of low microbial count and production.

With the number of isolated bacteria and fungi from the tested orange juice samples, it can be concluded that different bacterial and fungal species occur within fruits and materials used for the production of juice. This show that juices squeezed from fresh fruits for example sweet oranges can contain microorganisms which are potentially hazardous to public health.

Based on the processing methods and storage conditions applied in the study, it can be concluded that a safe microbial load and reduction in contamination of orange juice / fruit juices can be achieved by combination of the various processing methods: chemical preservations should be combined with pasteurisation, concentrated with carbonation under controlled microbiological environments such as freezer and refrigeration temperatures.

Considering the results of this present study the following recommendations are made: A combination of sodium metabisulphite (100ppm) and sodium benzoate (350ppm) (food preservatives) should be used as an antimicrobial and a flavouring agent and as adjuvant in the processing of orange juices in Nigerian markets. Good agricultural manufacturing practices and application of hot water and chemical sanitizers such as chlorine dioxide, ozone and peracetic acid should be used on surfaces of orange fruits prior to juice extraction in order to avoid and reduce microbial contaminants of fruit products. The Standards Organisation of Nigeria (SON) and National Agency for Food and Drug Administration and Control (NAFADAC) should ensure that the producers of locally manufactured commercial orange juice/fruit juices adhere strictly to specification for the maximum allowable limit of microorganisms for commercial fruit juices. They should carry out periodical inspection of production facilities and testing of products on the shelf, to ensure they conform to specified standards for food safety.

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