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Microbial and physicochemical properties of date jam with inclusion of apple and orange fruits

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Abstract

The microbial and physicochemical analysis of jam produced from date fruits enriched with apple and orange fruits at different proportions were investigated. Three (3) different blends of jam (coded as DOA, DO & DA) were produced with strawberry jam (SBJ) purchased from a store as reference jam. Sample SBJ (strawberry jam) has total plate count of 10.33×10^4 cfu/g, sample DOA has 55.00×10^4 cfu/g, sample DO had 13.67×10^4 cfu/g while sample DA has 17.67×10^4 cfu/g respectively. The fungi counts for all the samples were 8.33×10^4 cfu/g, 44.00×10^4 cfu/g, 13.33×10^4 cfu/g and 19.33×10^4 cfu/g. All the four samples showed positive (+ve) reactions in the biochemical tests indicating the presence of suspected glucose microbes such as bacillus spp., staphylococcus spp., pseudomonas spp. and streptococcus acid/gas forming bacteria. pH for the jam samples ranged from 3.14-4.52, total titratable acidity (TTA) varied from 0.60-0.68 while total soluble solid (TSS) indicating the brix level ranged from 43.00-51.00%. Colour analysis revealed no significant different (P≤0.05) between samples SBJ and DOA while samples DO and DA had significant differences.

Keywords: fruits, jam, microbial analyses, physicochemical properties, colour

Introduction

Date is the fruit obtain from date palm, which can be considered as an idea food that provides a wide range of essential nutrients with many potential health benefits. There are varieties of way in which date can be consumed. They are consumed as fresh (30%-40%) or in the dried form (60%-70%) at Rutal (semi-ripe) and Tamar (fully ripe) stages with little or no processing (Al-Hooti et.al, 1997)^[1]. Date can be taken with coffee, milk or yoghurt. When processed, they can be consumed as paste, syrup, pickles, jams and are also used in many bakeries or confectionaries product alongside with chocolate, coconut, honey, vinegar and others (Al-Hooti et.al, 1997; Besbes *et al*, 2009)^[1, 2].

Date consumption during the Muslim's holy month of fasting 'Ramadan' reaches its peak when it is commonly taken to break the fast. Date is said to contain antioxidant and antinuitagenic properties. Date and their aqueous extracts have demonstrated the free radical damage, antimotagenic and immunomodulatory activities (Vayalil, 2002)^[3].

The economic importance of date palm is due to its nutritionally valuable fruit which consists of 44.88% sugar, fat (0.2-0.5%) minerals such as potassium (2.5times more than bananas) calcium, magnesium and iron, protein 2.3-5.6%), dietary fibre (6.4-11.5%) as well as vitamin and amino acid (Al-Shahib and Marshall, 2003)^[4].

Jams are food made by boiling sugar to a thick consistency and is used in eating foods like bread, biscuits, pies e.t.c. (Merriam-Webster, 2000) ^[5]. Pamplona-Roga (2008) ^[6] stated that jams are prepared by combining sugar, pectin and citric or lemon juice with fruits and then heated the mixture till thick consistency is obtained. They are made from two main ingredients, fruit and sugar. The fruit has to contain plenty of pectin and acid for the jam to set properly.



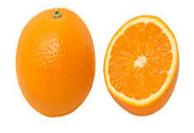


Fig 3: Orange

Apple juice is a fruit juice made by the maceration and pressing of apple fruit. The resulting expelled juice maybe further treated by enzymatic and centrifugal clarification to remove the starch and pectin while the orange juice is the liquid extract from the fruit.

Orange juice is the liquid extract from the fruit of the orange tree, produced by squeezing oranges. It comes in several different varieties. As well as variations in oranges used, some varieties include differing amounts of juice vesicles, known as pulp or juicy bits. How juicy these vesicles are depend upon many factors, such as species, variety and season

Materials and Method Sources of materials

Fresh matured date, orange and apple fruits were randomly picked and purchased from Sayedero a market in Ilaro metropolis, Yewa south local government area of Ogun state, southwest Nigeria. Commercial fruit jam, sugar (Dangote), fruit lam bottles were bought from a local store in Ilaro. All the materials were transported in a clean polythene bag to the laboratories of Department of Food Technology Federal Polytechnic, Ilaro for further processing and analyses.

Preparation of samples

The fruits were washed with clean water with damage ones removed prior to processing. Processing of orange, apple and date fruits for jam production.

Orange apple and date fruit were sorted out, washed, peeled, sliced and blended machine. After blending they were poured into pots with addition of sugar and cooked till they are done.

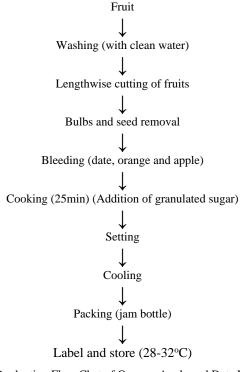


Fig 5: Production Flow Chat of Orange, Apple and Date Fruits for Jam

Table 1: Recipe used for Date jam with apple and orange

Jam ingredients	quantity
Fresh date	190g
Orange	30g
Apple	30g
Water	100ml (for bleeding)
Granulated sugar	100g

Table 2: Sample Ratios

Samples	Dates (%)	Orange (%)	Apple (%)
DOA	80	10	10
DO	90	10	-
SBJ	Strawberry	Jam	
DA	90	-	10



Fig 6: Date Jam: Orange



Fig 7: Date Jam: Apple

Analysis Microbiogical analysis i) Preparation of Media

The media used are Nutrient agar (N.A), and Potato Dextrose Agar (PDA). All media used were prepared according to the manufacturer's instructions. The mean counts of bacteria in colony forming fruits per gram of samples were determined.

ii) Method of preparation of Cultures.

The samples were serially diluted after maceration under aseptic conditions. The appropriate dilution (dilution 4) was inoculated on the two agar media. The bacteria were inoculated on Nutrient Agar for 24 -48 hours, fungi were inoculated on Potato Dextrose Agar (PDA) at $28 \pm 2^{\circ}$ C for 24 hours. Colonies on plates were counted and multiplied by the dilution factor.

iii) Identification of isolates

Inoculation was aseptically transferred from 10^{-4} into plates of respective media using a pour plate technique and it was gram stained. The isolates were purified by repeated pouring on their respective media. Bacteria plates were incubated at 37^{0} C for 24 hours and identified to the genus level by colony and cell morphology and biochemical tests according to at 25^{0} C for 25^{0} C for 72 hours. A 24 hours old culture was prepared from each plate for identification purposes, bacteria isolates were identified based on their cultural characteristics, Gram staining reaction and various identifications tests. The isolates were identified using their cultural and morphological characteristics were off vital importance in this process and were thus observed. Biochemical tests were also carried out.

iv) Procedure for Total Plate Count

All glass ware were sterilized in an area (the media nutrient agar) was prepared by weighing 7g and was dissolved in 250ml of distilled water, it was then sterilized in the oven and was allow to cooled to 45° c, the serial dilution of the four samples were carried out by pipetting 1ml of each of sample to already measured 9ml diluted water into a test tube labeled $10^{1} - 10^{5}$ and was covered with non-absorbent cotton wool to avoid contamination.

1ml from 10^4 of each sample was aseptically transferred into a sterile Petri dish for each plate was covered immediately. 20ml of thee cooled molten agar was poured into the Petri dish and rotated gently for thorough distribution of the inoculums through out of the medium and it was allow to solidify, the plate was inverted and incubated at 30° C for 48 hours.

v) Procedure for Yeast and Mould Count

Exactly 9ml of distilled water was pipette into 10 tubes and sterilized in an autoclave, then sterilized potato dextrose agar immediately before use. Pouring was done and Petri dishes swirled and allowed to solidify. The hardened agar was incubated at 37°C for 3-5 days in an inverted form and number of colony counted.

vi) Procedure for Gram Staining

On a clean slide, a smear of the organisms were made by dropping distilled water on canter of a clean slide. A loopful of bacteria colonies were transformed with a flame sterilized loop into the drop of water on the slide and then spread into a thin smear along the slide. The smear was allowed to air and fix with gentle heat by passing the slide over a Bunsen flame. The smear was flooded with crystal violet solution for 30-60 seconds and it was rinsed with water under a tap and Lugol's iodine was applied and allowed to act for 1minute.

It was decolorized with alcohol and left on the slide for 5-10 seconds and it was later rinsed under running tap water. The slide was then flood with a safranin solution and allowed to act for 2 minutes. It was then rinsed off under tap water and blot dry. It was examined using oil immersion, shape and colour of the bacteria all was noted and gram negative and gram positive bacteria were identified.

vii) Catalase Test

About 2ml of hydrogen peroxide was poured into test- tube and streaks of the isolate were introduced. Bubble notice when the organisms were introduced with, into the solution show that the organism is catalase negative.

viii) Oxidase Test

A few drop of ammodimethylaniline were dropped into a piece of filter paper in a Petri dish with a glass rod, some bacteria growth was smeared into the impregnated within 5 seconds for oxidize positive culture. A delayed reaction as recorded as negative (Seeley and Van Demark, 19 72)^[7].

ix) Coagulase Test (Slide screening test)

Two drops of coagulase plasma was placed in the centre of a slide. An inoculating lap was used to transfer in isolated colony from the streak plate and it was emulsified in the drop of the plasma. If the organism is coagulase positive, it will agglutinate, where as a coagulase negative organism will emulsify and produce a uniformly hazy suspension.

x) Sugar Fermentation (Sucrose and Glucose)

This is a test of the ability of an organism to ferment a particular sugar and cause a change in P^{H} of the medium (cad production) accompanied sometimes by the production of gas (e.g H₂, 1.2 from a peptone water culture, inoculation with wire loop into sugar both provided was made containing inverted Durham tube filled with the both, it was incubated at 37^{0} C for 24 hours. Acid production was indicated by a change in colour of the sugar both white gas was detected as an air space above the medium in the Durham tube.

Physicochemical Analysis

xi) Determination of P^H

The P^H of the samples was determined according to the method of AOAC, 2000)^[8]. 100 ml of the filtrate sample was measured using a P^H meter.

xii) Determination of Total Titratable Acidity (TTA)

10ml aliquots were pipetted and titrated against 0.1ml NAOH to phenolphthalein and point and the acidity was calculated as gram malic acid/100. (AOAC, 2000)^[8].

xiii) Determination of Total Soluble Solid (Brix %)

This was determined through the use of Abbe 60 refractometer corrected to 60°C according to Pearson (1981)^[9]. Before use, the refractometer was adjusted to zero reading using distilled water. Aliquot from the samples were placed on the prism surface of the refractometer and the total soluble solid directly as the sugar contents.

xiv) Determination of Colour

This was determined through the use of colorimeter. The colorimeter was calibrated with the provided sample. Then the readings of the samples were taken.

Result and Discussion

 Table 3: Showing the microbial analysis of jam produced from dates, orange and apple at different blends ratio

Sample Codes	Total count (cfu/g)	Fungi Count (cfu/g)
DOA	17.67 ±4.51 ^b	19.33±4.51 ^b
DO	13.67 ±3.51 ^{ab}	13.33 ±5.51 ^a
SBJ	55.00 ±13.25°	44.00±4.58°
DA	$10.33 \pm 0.8^{\text{d}}$	8.33 ±3.51 ^a

Keys: DOA= Date Jam (80% Date, 10% Apple and 10% Orange) DO = Date Jam (90% Date and 10% Orange) SBJ = Strawberry Jam Purchased from food store

DA = Date Jam (90% Date and 10% Apple)

DA – Date Jain (90% Date and 10% Apple)

Tables 4: Biochemical tests for bacterial isolation of jams

 produced from date, orange and apple at different blends (ratios)

	Suspected Glucose	Shape	Gram Staining	Catalase	Oxidase	Coagulase
	Bacillus sp	Rod	+ve	+ve	Acid/gas	Acid/gas
	Staphylococcu	Cocci	+ve	+ve	Acid/gas	Acid/gas
Γ	Pseudomonas	Rod	+ve	+ve	Acid/gas	Acid/gas
	Streptococcus	Cocci	+ve	+ve	Acid/gas	Acid/gas

 Table 5: Physicochemical properties of jam produced from dates, orange and apple at different blends (ratios)

Sample codes	pН	(TTA)	TTS (Brix %)
DOA	3.30 ±2.03 ^b	0.60 ± 0.00^{b}	45.0 ±0.00 ^a
DO	3.42 ±0.02 ^a	0.67 ± 0.00^{a}	49.0 ±0.00 ^b
STJ	4.52 ±0.03 ^d	0.68 ± 0.00^{a}	51.0 ± 0.00^{b}
DA	3.14 ±0.02 ^a	0.60 ±0.00 b	43.0 ±0.00 ^a

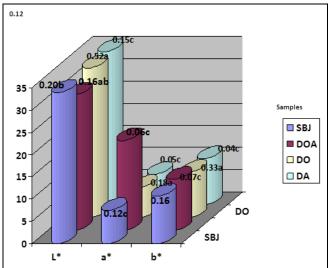
Values were means standard deviation of duplicate Keys: DOA= Date Jam (80% Date, 10% Apple and 10% Orange) DO = Date Jam (90% Date and 10% Orange)

SBJ = Strawberry Jam Purchased from food store

DA = Date Jam (90% Date and 10% Apple)

TTA= Total titratable acidity

TTS=Total Solid



Keys: DOA= Date Jam (80% Date, 10% Apple and 10% Orange) DO = Date Jam (90% Date and 10% Orange)

SBJ = Strawberry Jam Purchased from food store

- DA = Date Jam (90% Date and 10% Apple)
- L*= Lightness
- a* = Redness
- b*= Yellowness

Fig 6: Physicochemical properties (colour) of jam produced from dates, orange and apple at different blends (ratios)

Discussion

Microbial Analysis

The total plate count for all the samples including the reference sample (SBJ) range from 10.33×104-55.00×104 cfu/g reflecting the significant differences (P≤0.05) among samples. Sample SBJ has the least total plate count while sample DOA (Date: Orange: Apple). The fungi counts revealed 8.33×10⁴, 44.00×10⁴, 13.33×10⁴ and 19.33×10⁴ cfu/g for sample SBJ, DOA, DO and DA respectively. Also there were significant differences with ($P \le 0.05$) in the fungi count. However, both the total plate counts and the fungi counts do not exceed the standard (×106cfu/g) (ICMSF, 2002)^[10]. The reduction in the microbial levels may be due to the intense heat application involved in jam production as well as the high pH and the sugar content of the product. All this jam samples showed positive reactions in the biochemical test indicating the presence of suspected glucose forming microbes such as bacillus spp., staphylococcus spp., pseudomonas spp. and streptococcus spp. which are all acid/gas forming bacteria.

Physicochemical Analysis

The pH is one of the most important factors that must be

monitored and controlled in jam production for optimum gel condition. pH values of 4.52, 3.30, 3.42 and 3.14 were obtained for samples SBJ, DOA, DO and DA respectively, showing significant differences among samples strawberry jam (SBJ) has the least pH value while date jam (DA) has the highest value. These values are within the prescribed limit of FAO (2006)^[11]. The result also is similar to those recorded in a previous work (Devotha, 2015) ^[12] for jam produced with jackfruit. Low pH in foods generally prevents the microbial growth. Total titratable acidity varied from 0.60-0.68 for all sample under consideration. The result obtained is in agreement with those reported by Devotha (2015)^[12] for jam produced with jackfruits. The importance of high acidity in food products shows that such foods can be stored for sometimes before spoilage or deterioration set in. Acidity is also useful to correct acid/sugar ratio need in jam production. Sample coded SBJ (strawberry jam) has the highest brix value of 51.0% while sample coded DO (Date and Orange jam) had the least brix value of 43.0%. According to India standard, the total soluble solids for jam should not be less than 68% (BIS, 1993)^[13]. Sugar generally contributes to soluble solids in jam production an effect that is essential for the physical and chemical properties, thus proving the body and month feels improves appearance (colour and shine) and makes gelation of pectin possible (Hyvönen and Törmä, 1983)^[14].

Colour Analysis

The results of the colour analysis of jam are as shown in chart above shows no significant difference (P \leq 0.05) in lightness between sample SBJ and DOA while sample DO and DA has significant differences high. In terms of the redness of the sample, there are significant differences (P \leq 0.05) between sample SBJ and DOA (i.e strawberry jam and date: orange: apple jam). Sample DO and DA has no significant difference (P \leq 0.05) as the showed values which are very close i.e. 6.88 and 6.74 respectively. The yellowness of the four samples showed significant differences (P \leq 0.05) among them probably due to colour of each fruit and this is similar to a previous work reported by Al-baki *et al.* (2002)^[15] for colour analysis on fruits.

Conclusion

It is concluded that date fruit which is an underutilized fruit in combination with other notable fruit can be used to produce jam which are nutritionally and microbiologically safe for human consumption. It is also an opportunity to explore the possibility of converting the fruit to other products such as jam, thereby reducing post-harvest losses of fruit.

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