

A study on the microbiological status of few samples of packed fruit juice

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ABSTRACT

In the present study twenty samples, each 10 of 2 different Packets fruit juice (A&B) marketed in Karimnagar city, Andhra Pradesh was assessed by using the parameters: the number of microorganisms, types of microorganisms, the presence of coliforms, the yeast and mold content of the fruit juice samples. Different packeted fruit juice samples were collected and the samples were surface sterilized with IPA. Then the packets were thoroughly shaken and mixed and then 1 ml was taken into the sterile pipette and inoculated in to the desirable medium. All the samples from different pocketed fruit juice showed the presence of bacteria, yeast, mold, and coliforms being absent.

Key words: Packet Fruit Juice, Microbiological status.

INTRODUCTION

Two different pocketed fruit juice marketed in retail shops in Karimnagar city of Andhra Pradesh were designed A & B in this study. All the two pocketed fruit juice carried code numbers. These two pocketed fruit juice are available in the most of the outlets. These two pocketed fruit juice contain vegetable hologram symbol, and contain ingredients like water, juice, permitted preservatives, antioxidants, citric acid. This was also certified by the FPO stored in refrigerators at the temperatures of approximately 2-3°C.

MATERIAL AND METHODS

Different packet fruit juice samples were collected from different retail outlets and all were brought into the laboratory. All the samples were surface sterilized with IPA (70%), and then the packets were thoroughly shaken, followed by thorough mixing the packet sample was withdrawn aseptically and then 1 ml of the sample was collected by sterile pipette and inoculated in to desirable medium.

Isolation and identification of bacterial isolates: From the standard plate count agar plates, colonies were streaked on nutrient agar plates and kept overnight inoculation. After inoculation period the colony from nutrient agar plates were picked

up and inoculated in to sterile peptone water and incubated for 4-6 hours. This was used for microscopic and various biochemical identification tests.

Microscopic examination: Using gram staining method in order to identify the morphology of bacteria stained the colony obtained by the plate count technique. The smear was prepared in a grease free slide using the individual colony on the medium. The smear was flooded with crystal violet solution and allowed to stand for one minute. Later it was washed with water and flooded with gram iodine solution and allowed to stand for two minutes. Then it was drained and decolourized with 95% ethanol, followed by washing gently in running tap water. Then the slide was stained with a counter stain called safranin for 30 minutes. After drying the stained smear was observed under microscope to identify the organisms to place them in their individual reaction to certain stains as gram-positive or gram-negative cocci or rods.

Lactophenol cotton blue mount: A small portion of fungal cultures were taken and suspended in lactophenol cotton blue solution on a clean slide, tested well and covered with cover slip without air bubbles and observed under low power objective. Motility is used to identify whether the organism obtained is motile or non-motile.

The hanging drop technique was followed to observe the motility of the bacteria. A drop of suspension of culture was placed at the centre of the cover slip, and placed in an inverted position over a cavity slide, so that the drop is hanging over the cavity. The edge of the drop was observed under low power lens of a microscope where the movements of the organism can be seen.

RESULTS AND DISCUSSION

The microbial analysis of pocketed fruit juice A and B was detailed in tables-1 & 2. The bacterial flora of pocketed fruit juice A and B was detailed in tables-3 & 4. The microbial flora and percentage of bacterial flora of pocketed fruit juice A and B was detailed in tables-5 and 6. Results for the morphological, cultural characteristics of the isolates, and results for the confirmatory tests of the bacterial isolates were detailed in tables-7 and 8 and biochemical tests resulted was detailed in table-9.

Table 1: Microbial analysis of pocketed fruit juice A.

Sr. No.	Total platelet count/ml	Yeast count	Mold count	Coliforms count
1	11	04	-	-
2	21	-	-	-
3	08	-	-	-
4	16	06	02	-
5	14	-	-	-
6	12	01	01	-
7	15	-	-	-
8	19	-	-	-
9	10	-	-	-
10	13	05	03	-

Table 2: Microbial analysis of pocketed fruit juice B.

Sr. No.	Total platelet count/ml	Yeast count	Mold count	Coliforms count
1	08	03	-	-
2	12	-	-	-
3	19	06	02	-
4	13	-	-	-
5	06	02	03	-
6	14	-	-	-
7	17	01	-	-
8	09	-	-	-
9	15	-	-	-
10	18	05	03	-

Table 3: Bacterial flora of packeted fruit juice A.

Sl. No.	Isolated Occurrence	Number of Isolates	Percentage of
1	<i>Staphylococcus aureus</i>	5	38
2	<i>Bacillus cereus</i>	6	62
3	<i>Psuedomonas aeruginosa</i>	1	07

Table 4: Bacterial flora of packeted fruit juice b.

Sl. No.	Isolated Occurrence	Number of Isolates	Percentage of
1	<i>Staphylococcus aureus</i>	3	23
2	<i>Bacillus cereus</i>	9	69
3	<i>Psuedomonas aeruginosa</i>	1	07

Table 5: average count of microbial flora

Sl. No.	Packed Fruit Juice	Bacterial Count	Yeast Count	Mold Count
1	A	14	1.5	1.6
2	B	13	1.4	0.7

Table-6: percentage of different bacterial flora

Sl. No.	Packed Fruit Juice	<i>Bacillus cereus</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
1	A	62%	38%	-
2	B	69%	23%	7%

TABLE-7: MORPHOLOGICAL CULTURAL CHARACTERISTICS OF THE ISOLATES

Microscopic Examination			Colony Morphology	Sugar Test	Organisms
Gram-Strain	Spore-Strain	Motility-Strain	Nutrient Sugar- Blood Sugar	G S L M	
G+ve	+	+	White rhizoid – B-haemolysis	+ - + -	<i>Baccillus cereus</i>
G-ve	-	+	Bluish Green –B-haemolysis	+ - - -	<i>Pseudomonas</i>
G+ve	+	-	Golden yellow –B-haemolysis	+ + + +	<i>Staphylococcus</i>

Abbreviations:- G= Glucose: S= Sucrose: L= Lactose: M= Mannitol

Table- 8: The confirmatory tests of the bacterial isolates, grown in selective media.

Organisms	BCA	CA	MSA
<i>Bacillus cereus</i>	Present	Absent	Absent
<i>Pseudomonas aeruginosa</i>	Absent	Present	Absent
<i>Staphylococcus aureus</i>	Absent	Absent	Present

Abbreviations: BCA=Bacillus cereus agar; CA= Citrimide agar; MSA=Mantol sugar

Table 9: Biochemical tests of the bacterial isolates.

Organisms	CAT	OXI	I	VP	CIT	URE	COA
<i>Bacillus cereus</i>	-	+	-	-	-	-	-
<i>Pseudomonas aeruginosa</i>	+	+	-	-	+	-	-
<i>Staphylococcus aureus</i>	+	-	-	+	-	-	+

Abbreviations: CAT= Catelase; OXI= Oxidase; I= Indole; MR= Methylred; VP= Voges proskauer; CIT= citrate; URE= Urease; COA= Coagulase test;

10 samples from packeted fruit juice A has been studied for hire microbial flora in table-1. The total plate count varied from 18 to 21 with an average of 14 cfu/ml. Out of 10 samples processed for yeast and mold count 40% showed yeast and mold growth with a range of 1 to 6 yeast and 1 to 3 mold growth. None of the samples showed presence of coli forms.

10 samples from packeted fruit juice B has been studied for microbial flora in table-2. The total plate count varied from 8 to 19 with an average of 13 cfu/ml. Out of 10 samples processed for yeast and mold count 50% showed yeasts and mold growth with a range of 1 to 5 yeast and 2 to 3 mold growth. None of the samples showed presence of coli forms.

Sugar has been recognized as the source of endospores of most of the *Bacillus* species, which find their way in the final drink product. This may be the reason for the presence of *Bacillus* in large percentage of the soft drinks. These bacterial species may survive in the usual hot fill process that is given to commercial juices.

In the present study all the brands with organisms are not significant when considering the stability of the juice as spore forming bacteria predominated. However, such results usually raise

a doubt as to the quality of fruit used. In the present study all the samples of the four brands as showed *Bacillus* growth. *Bacillus* presence is found high in brand B. *Staphylococcus aureus* may be associated with the skin, nose and throat of healthy individuals. It is used as an indicator of general hygiene to good handling procedures.

In the present study all the brands showed the presence of *Staphylococcus aureus*, which are fairly good in number. This may be due to contamination from the workers during preparation may be the reason for the presence of *Staphylococcus* in the soft drinks. *Pseudomonas* species are ubiquitous in the environment, water and significant in food spoilage, particularly in chilled foods. Higher levels more than 10⁷ cfu/ml may result in the flavours and off odours.

The presence of small number of the coliforms is not a proof that there has been contaminated during the production. Molds are generally considered to be the least important group of microorganisms causing spoilage in fruit juices. The milling and pressing equipment that is not cleaned daily is the major source of inoculation. If fruit juices are not kept in cold storage, it may contaminate with yeast cells.

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