

BACTERIOLOGICAL LOAD ASSESSMENT OF JUICE SOLD IN CAFETERIA AND HOTELS IN ARBA MINCH TOWN, GAMO GOFA, SOUTHERN ETHIOPIA.

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Abstract:-

Fruit juices are the specialized drinks which are rich in vitamins and nutrients that can help prevent common birth defects as well as cancer and also contain enzymes that are essential for digestive process. The study was performed to evaluate the microbiological quality of locally processed juices such as mango and avocado juices from some selected hotels and cafeteria found in Arba Minch town. Four mango and Avocado, total 8 juice samples from two hotels and cafeterias were collected and their microbial load was analyzed in Microbiology laboratory on appropriate media by using serial dilution techniques followed by spread plate techniques. The highest colonies were observed from mango juice sample from cafeteria two and the lowest colony from hotel one are 94 colonies and 0 colony respectively. The mean bacterial count of avocado juice sample was 49500 cfu/ml which was relatively higher than the mean colonies of mango's juice sample was 15420 cfu/ml. The total mean of bacterial colonies were 63750 cfu/ml from nutrient agar media, 16250 cfu/ml from Manito salt agar and 23250 cfu/ml from Macconkey in case of cafeteria as well as 41250 cfu/ml from nutrient agar media, 22250 cfu/ml from Manito salt agar and 28000 cfu/ml from Macconkey in case of hotels. Their for, juice venders that produce unpasteurized mango avocado juices would be recommended that preventative measures through food safety control strategies is important.

Keywords: - Fruit juice, Health, risk, hygiene, Bacterial count

1. INTRODUCTION

Freshly squeezed juices are simply prepared by extracting the liquid and pulp of mature fruit usually by mechanical means or blenders. Prior preparation of fruit to avoid bitterness of skin or to remove target stone such as mango, avocado and pine apple followed by separation of juices and pulp by blender, final juice ready for consumption. However, it is well known fact that food serves as very good medium for growth of microorganism especially when the principles of hygiene and sanitation are not met the food become contaminated by pathogens from human or from the environment during production, processing or preparation. Especially local preparation of fruit juice has no processes that reduce pathogen levels, if contaminated, such as microbe killing step (Melbourne, 2006).

Ethiopia, most of the fruit juices being served in area had higher microbial load than the specification set for fruit juices in some parts of the world. As the products could be the cause of health problems and potential vehicle of food borne outbreaks, high levels of works hygiene should be enforced and the use of disinfectant better practiced to improve the microbial quality, safety and shelf life of the final products (Ketema *et al.*, 2008).

Pathogenic organisms can enter to fruits through damaged surfaces, such as punctures, wounds, cuts. This damage can occurs during maturation or during harvesting and processing; pathogen that has become internalized with in a fruit must be able to survive in the product until it reaches the consumer in order to become public health hazard. Most fruit juices are sufficiently acidic to inhibit the growth of public pathogenic organisms (Melbourne, 2005)

In addition, during the process contamination from raw materials; equipment or food handlers, the pathogen could be easily transferred to the final products of pathogen such as *staphylococcus aureus*, *salmonellae*, *shigella*, *E.coli* O157:H7, etc were present in freshly fruit be improver washing of the fruits, use of un hygienic water for dilution, prolonged preservation without refrigeration; unhygienic environment for juices preparation and poor handling of properly preferred juices (Lewis *et al.*, 2006) food borne or waterborne microbial pathogens are leading causes of illnesses in developing countries, killing an estimated 1.9 million peoples annually at the global levels. Even in the developed countries, an estimated. one third or the population is affected by micro biological food borne disease each year (Andargie *et al.*, 2008).there are reports of food borne illness associated with the consumption of fruit juices of the several places of India and elsewhere (Sospedra *et al.*, 2012). The most commonly reported bacterial genera include Acetobacter, Alicyclobacillus, Bacillus, Gluconobacter, Lactobacillus, Leuconostoc, Zymomonas, and Zymobacter. Among yeast: Picha, Candida, Saccharomyces, and Rhodotorula are commonly encountered genera responsible for spoilage of juice (Ghenghesh *et al.*, 2005).

The most likely cause of the contamination of fruit coming in contact with animal feces or water, workers containers or processing equipment contaminated with animal feces; cattle, deer and sheep are the most common reserves for pathogen ,but usually don't show symptoms themselves ,birds ,rodent, insects and poor hygiene may also contributed to the contamination. One contaminated price of fruit could affect an enter batch of juice or cider (FDA, 1999).

Poor handling and processing of fresh fruit juices are some of the main sources or cause of food borne associated illness to the community who lives in developing countries. In most case a number of pathogenic organisms are located and identified from locally prepared fruit juices. According to study conducted in Dhaka, Bangladesh, the total viable count of samples ranged from 3.00×10^2 to 9.60×10^8 out of 114 freshly prepared fruit juices samples collected, 113 samples (99%) showed the presence of coli form and E.coli. The others bacteria like B.cereus, staphylococcus, salmonella, streptococcus were found in 64.91%, 6.14%, 7.89% and 5.26% of the tested samples, respectively. The number and types of microorganism recovered from the freshly squeezed fruit juices made them unsafe for drinking. It was concluded that due to unhygienic fruit handling in the unsanitary environmental conditions under which the vendors operate the juices becomes contaminated with harm full bacteria (Shaker *et al.*, 2009)

1.2. Statement of the Problem

Bacteria have significant effect for quality of squeezed fruit juices which are sold in cafeteria and hotels in Arba Minch town. It is difficult to get enough and qualified juices from any hotels and cafeteria without any problem for consumption. Because, due to any no care for the preparation of juice and lack of fruit storage practice, in almost all hotels and cafeteria, the fruit juices and other fruit products would be contaminated by pathogenic microbes especially bacteria and fungi that makes fruit juices unacceptable for human's consumption. So, taking care for preparation of juice as well as fruit storing on temporal cleaned area and at optimum temperature is very important for consumers.

1.3. Objectives of the Study

1.3.1. General Objective

The general objective of the study was to assess bacteriological quality, safety of freshly squeezed and unpasteurized fruit juices in cafeteria and hotels which are sold in Arba Minch

1.3.2. Specific Objectives

- To assess bacteriological quality of freshly squeezed fruit juices.
- To determine bacteriological load of juices from both Hotels of Cafeteria
- To compare bacteriological features of juice from Hotel and Cafeteria

- To determine which type of juice can easily contaminated by microbes

1.4. Significance of the Study

The significance of this study would be information sources for fruit juice producers and users peoples in order to get some knowledge about importance, the sources of contamination which affect the quality of locally prepared fruit juices as well as the way that create the favorite conditions for the growth of pathogenic bacteria in fruit juices. The study also help to create awareness who to solve the problem have occurred in the study area or hotels and cafeteria. It also important for the hotel's and cafeteria's managers to produce the juices in proper way and to know the amount of prepared juices must be matched with his/her customer in order to minimize the expiration rate. It also help to provide information for further study and to know which type of juice can easily contaminated by food born disease

2. Matherials and methods

2.1. Description of the Study Area

The study will conducted in Arba Minch town which is found in Gamo Gofa zone in southern part of Ethiopia. Arba Minch is a city separate woreda in southern Ethiopia. It located in the Gamo Gofa of the Southern Nations, Nationalities, and Peoples Region at about 500 kilo meters south of Addis Ababa, at an elevation of 1285 meters above sea level. It is the largest town in Gamo Gofa zone and the second town in SNNPR next to Awassa. Arba Minch received its name for the abundant local springs which produce a groundwater forest. Located at the base of the western side of the Great Rift Valley, Arba Minch consists of the uptown administrative centre of Shecha and 4 kilometers away the downtown commercial and residential areas of sikela, which are connected by a paved road. On the eastern side of sikela is the gate to Nechisar National park, which covers the isthmus between Lake Abaya to the north and Lake Chamo to the south. It is known as a source for fruit, including mango, banana, orange, apple, guava and pineapple as well as known for its fish farms. (Philip, 2002).

2.2. Study Design

Cross sectional study was used to evaluate the bacteriological load as well as quality assessment of locally prepared fresh fruit juices in Arba Minch town.

2.3. Sources of Samples

The sources of the samples were unpasteurized juices from selected cafeterias and hotels found in Arba Minch town. Two most commonly used unpasteurized juices (Mango and Avocado) were collected from two Hotels and Cafeterias found in the town.

3.5.2. Sample techniques and Sample size

Random sample technique based on accessibility of hotels and cafeteria around Abaya campus and their cost to Purchas juice sample was used to collect juice sample. Sample size was determined by convenience based on time and budget. Accordingly, two hotels and two cafeterias were assessed. Totally eight samples were collected (four mango juice samples and four avocado juice samples) from those two selected cafeteria and hotels.

2.6. Sample transportation and laboratory procedure

3.6.1. Sample transporting

The all purchased juice samples were transferred in to sterilized test tubes which were taken from Arba Minch university microbiology laboratory class and labeled with their names and their site of collection, and then taken to micro biology laboratory of Arba Minch University for analysis.

2.6.2. Sample Processing

A total of 100ml of each sample was taken and transferred with a sterilized micro pipette to the different test tubes containing 10ml of double distilled sterilized water to made 10^{-1} dilution. In such way, dilution up to 10^{-3} was made. After that the 1ml of each sample from the test tubes with 10^{-3} dilution was taken by sterilized micro pipette and inoculated to different pettier dishes containing three different medium (Nutrient agar, Manito salt agar and Macconkey), then labeled each dishes with the type of sample inoculated on it and the site from which the sample collected. Neatly the cultured medium were kept in incubator for 24 hours at temperature of 35°C and all colonies on the each dishes were counted using a colony counter and then the counted colonies were converted to number of colonies in original sample by the following formula.

$$\text{No. of bacteria colony in original sample} = \text{Number of colony counted} \times \frac{1}{df} \times V \text{ (ml)}$$

Where: df = Dilution factor, V = Volume in ml

2.7. Data analysis

The data was analyzed using descriptive statistical parameters like tables and graphs that are observed from the experiment.

3. RESULT AND DISCUSSION

3.1. Results

After 24 hours late, the numerous of bacteria can be grown on each different medium. The relatively high loads of different bacterial groups were observed as the form of colonies from the fruit juices examined in this study and the total numbers of bacterial count in original juice samples could be calculated by the given formula was calculated as following.

$$\text{No. of bacterial colony in Original Sample} = \text{No. of colony counted} \times \frac{1}{df} \times V(\text{ml})$$

Where: df = dilution factor, V = volume (ml).

Table 1: Bacterial colonies grown on different medium

Site of collection	Type of juice sample	Type of media	No. colonies	No. of bacteria in original sample
Hotel 1	Mango	Nutrient agar	8 colonies	8×10^3 cfu/ml
		Manito salt agar	6 colonies	6×10^3 cfu/ml
		Macconkey	0 colony	0
	Avocado	Nutrient agar	70 colonies	7×10^3 cfu/ml
		Manito salt agar	60 colonies	60×10^3 cfu/ml
		Macconkey	47 colonies	47×10^3 cfu/ml
Hotel2	Mango	Nutrient agar	4 colonies	4×10^3 cfu/ml
		Manito salt agar	2 colonies	2×10^3 cfu/ml
		Macconkey	1 colony	1×10^3 cfu/ml
	Avocado	Nutrient agar	83 colonies	83×10^3 cfu/ml
		Manito salt agar	21 colonies	21×10^3 cfu/ml
		Macconkey	64 colonies	64×10^3 cfu/ml
Cafeteria1	Mango	Nutrient agar	29 colonies	29×10^3 cfu/ml
		Manito salt agar	7 colonies	7×10^3 cfu/ml
		Macconkey	2 colonies	2×10^3 cfu/ml
	Avocado	Nutrient agar	79 colonies	79×10^3 cfu/ml
		Manito salt agar	13 colonies	13×10^3 cfu/ml
		Macconkey	38 colonies	38×10^3 cfu/ml
Cafeteria2	Mango	Nutrient agar	94 colonies	94×10^3 cfu/ml
		Manito salt agar	28 colonies	28×10^3 cfu/ml
		Macconkey	4 colonies	4×10^3 cfu/ml
	Avocado	Nutrient agar	53 colonies	53×10^3 cfu/ml
		Manito salt agar	17 colonies	17×10^3 cfu/ml
		Macconkey	49 colonies	49×10^3 cfu/ml

Table 1 above Shows that all the recorded results of bacterial colonies and the total number of bacteria grow from original sample on three different types of medium from the experiment. More colonies was observed on samples from cafeteria two which was around 94×10^3 cfu/ml while less colonies was observed on samples from hotel one which was 0 colony.

3.2. DISCUSSION

The microbial contamination present in commercially available and freshly prepared mango and avocado juices samples collected from different localities of Arba Minch town from nechisar kifle ketema around Nechisar campus and from sikela at the center around Gamo square. The results showed the presence of too many pathogenic bacteria in commercially available mango and avocado juices which clearly demonstrates the unhygienic conditions of processing plants. In most

localities, the freshly squeezed mango and avocado juices were hygienically very poor as bacterial loads on the whole were abnormally high.

According to the research conducted in university of Valencia in Spain, poor handling of fruits and insufficient cleaning of juicing equipment's in the food service setting can stimulate bacterial contamination that can results in food-borne illness outbreak. Juicing machines have a large surface area and many holes and cavities that can promote microbial contamination, which is picked up by the juice as it is being prepared (Sospedra *et al.*, 2012).

From above recorded results the mean bacterial count in fresh juice samples of cafeteria was 34420cfu/ml. The total bacterial count from Nutrient agar media of two different juice samples was 63750cfu/ml with range from 29×10^3 cfu/ml to 94×10^3 cfu/ml. Similarly, we found mean Gram positive bacteria (staphylococcus) count from Manito salt agar media was 16250 cfu/ml with range from 7×10^3 cfu/ml to 28×10^3 cfu/ml. In case of total Gram negative bacterial count from Macconkey, we found the mean count was 23250cfu/ml with the range from 2×10^3 cfu/ml to 49×10^3 cfu/ml. While, the mean total bacterial counts in fresh juice samples of Hotels was 30500cfu/ml. The total mean of bacterial count from Nutrient agar media was 41250 cfu/ml with range from 4×10^3 cfu/ml to 83×10^3 cfu/ml. In case of Manito salt agar media, we found mean gram positive bacterial count was 22250cfu/ml with range from 2×10^3 cfu/ml to 60×10^3 cfu/ml. Also from MacConkey, we found the mean gram negative bacterial count was 28000 cfu/ml with range from 0 to 64×10^3 cfu/ml.

Based on above idea briefly explained that, the juices which were produced from cafeteria are more contaminated than the juices from hotels. This may be due to lack of fruit storage practice and safety from handlers, lack of any care for raw materials needed for juice production, lack of cleanity for storage area of produced juice products, improper follow juice making procedure, etc. Fruit have its temporary storage sites but most of the juice producers had no special storage sites for their fruits. The reasons for proliferation of microorganisms in fruit juices could also be attributed to the fact that the most juice producers lacked special training in food hygiene and safety. The total average numbers of colonies counted in case of both cafeteria and hotels from each different type of media, on the nutrient agar the high numbers of colonies are counted. Nextly from Macconkey and the less from Manito salt agar when compared to each others. This is may be different in their nutritional value and due to their selectivity to growth of some bacteria. So, according to this we can give some opinion in own that, most of freshly squeezed fruit juices which sold in Arba Minch town's cafeteria and hotels can contain less number of gram positive bacteria, but they present in all juice types according to our research. Because gram positive bacteria could grow from all juice samples that were collected from different sites in the town.

When we compare the total average of bacteria which grow from Avocado juice with Mango juice, the more bacteria can grow on Avocado juice than Mango juice. Because, the total mean of bacteria from avocado juice is 4.950×10^4 cfu/ml and the total mean of bacteria from mango juice is 1.542×10^4 cfu/ml. This is indicates that Avocado juice can easily contaminated. According to the study conducted on microbiological safety of fruit juices served in cafes/restaurants in Jimma town, Southwest Ethiopia from February-2005 up to July-2006, the mean counts of staphylococci, yeasts and molds (generally pathogenic microbes) were the highest in avocado juices in all cases (Tsige K. *et al.*, 2008), which makes it similar with our study conducted in Arba Minch town on mango and avocado juices. So from this generalized that, the Avocado juices were sensitive to contamination and the good medium for microbial growth under poor hygienic conditions and present many more pathogenic microorganisms that cause food borne associated disease rather than the other types of juices due to its high nutrient contents. Unlike to our study, the study was conducted for colony counted as well as for identification of types of microbial loads from each juice samples.

The products with high water activity possess good amount of unbound water molecule that supports growth and survival of microorganisms. However, the low acidity (i.e. higher P^H) and viscosity of avocado, besides its nutrient content, makes it good medium for growth of microorganisms. Some members of Gram positive bacteria (staphylococcus species) were encountered in all the juice samples rather than Gram negative bacteria (for example:-in mango juice sample from Hotel-1 on Macconkey media, the bacterial colony counted is zero). This result shows there are no gram negative bacteria present in mango juice which produced from this hotel. The presence of gram negative bacteria (staphylococcus species) in almost all the juice samples can be attributed to contamination via handling. This is may be due to poor personal and domestic hygiene indicating lack of knowledge of hygienic practices and safety of food products (Tambekar *et al.*, 2009; Bello *et al.*, 2013). On the basis of the gulf standards, it is clear that the colony counts of the microbial groups in our fruit juice samples exceeded the standard by considerable margin. These high counts, however may posse hazard to the health of consumers especially if pathogenic species are present in the fruit juices to be consumed.

4. CONCLUSION AND RECOMANDATION

4.1. Conclusion

Based on the presence of uncountable bacteria in fresh mango and avocado juices, it is concluded that juices in certain areas inside the Arba Minch town are highly impacted and unfit for human consumption. It is agreed that such type of contamination is the result of bad quality water used for dilution of juices, unhygienic surroundings and locations of stalls by the side of a busy road with vehicular traffic or by the side of waste disposal and overcrowding, poor practice of fruit storage and juicing machine as well as poor hygienic practice from vendors. Generally, this study shows us consumption

of fruit juices could have both positive and negative effect on the part of consumers. The juices produced under hygienic condition could have positive effect in enhancing consumer's health through inhibition of breast cancer, congestive heart failure, and urinary tract infection. While, in the absence of good manufacturing practices, the nutritional richness of fruit juices makes the product good medium for microbial growth, vehicle of food borne pathogens and associated disease.

4.2. Recommendation

The practice of consuming fruit and vegetable juices cannot be stopped on unhygienic grounds, and juice makers cannot also be prohibited from selling such items, since it is a source of their livelihood. However, to get health after consumption of fruit juice and to produce health juices for consumers the following ideas are recommended.

- Health agencies must be adopt measures to educate the vendors on food safety and hygienic practices.
- Regular monitoring of the quality of fruit and vegetable juices for human consumption must be enforced.
- There is need to educate the juice makers and retailers on the hazards associated with the cultivation of non-chalant attitudes to hygienic processing, display and packing of these juices.
- The people must be avoid consuming unpasteurized juices sold around road sides, they recommended to purchase and consuming only juices that has been pasteurized.
- Before manufacturing juice, the manufacturer must be take care for fruit storage practice and must be select the clean area and conditions in order to produce health and pasteurized juice for consumers.
- During juice processing the producers must be keeping their safety by washing their hands before handling the fruits and other necessary materials needed in fruit juice production and they must be covering their hairs.
- The juicing machines are must be cleaned and disinfected properly.
- Further studies must be conducted for identification bacteria to species level to set standards.

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