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Microbial Contamination Of Street Vended Fresh Fruit Juices Sold In Ranchi City.

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Abstract – Introduction -*Juices are prepared by mechanical squeezing of the pulp of fresh fruits or vegetables without application of heat. Improper washing of fruits before cutting[16] , inadequate hygiene maintenance by the juice makers, purchase of fruits from open markets having polluted environment and swarming houseflies and fruit flies, storage of the fruits on dirty dust filled shelves at room temperature and not in refrigerators, use of improperly cleaned utensils[12] and tumblers for processing and serving respectively are some of the most important factors which support microbial contamination of the fruit juices.* ***Aims and objectives -*** *Evaluating the quality and microbiological safety of fresh fruit juices vended on various streets of Ranchi, Jharkhand.* ***Materials and methods–*** *Fresh fruit juices were collected from 30 different street vendors across Ranchi city and inoculated on 6 different culture medias by spread plate technique.* ***Results*** *– All the samples collected were found to be contaminated and the organisms isolated were Enterobacter spp., Klebsiella spp., Pantoea agglomerans, Leclercia adecarboxylata, Lactobacillus spp., S.aureus, Pseudomonas spp., Proteus spp., Candida spp. etc.* ***Discussion*** *- Fruit juices are good medium for microbial growth. Hence, a number of organisms can flourish in them easily.* ***Conclusion*** *– Regular monitoring of the quality of street vended fresh fruit juices must be carried out.*

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I. Introduction

Fresh fruit and vegetable juices are an important part of modern day diet in many parts of the world as they are rich source of vitamins , minerals[2] and other phytochemicals which are of health benefits[9]. Juices improve blood lipid profiles in people affected by hypercholesterolemia and enhance consumers health through inhibition of cancers[2], diabetes[2], congestive heart failure , urinary tract infection[19]. Vitamin C is naturally present in juices which are essential for the body to form collagen, cartilage, muscle and blood vessels. It also helps in the absorption of iron[2]. Unpasteurized juices are preferred by people because of the fresh flavour. Moreover, during summers, juices garnished with ice cubes become the most preferred and easily available thirst quenchers for people on day to day basis. But these ice cubes[8] or water[4] used for diluting the juices act as a significant source of contamination for the juices being served. Pathogenic organisms can enter fruits through damaged surfaces, such as splits or punctures that happen especially when the ripened fruits fall down from the tree, on the ground or soil[2] that was already contaminated. The main ingredients of fruit juices are pulp, water, sugar etc. which may carry some microbial load with them[18]. Most fruits contain bacterial counts upto 1.0×10^5 CFU/cm^2 on their surface[8][22] [21]. The pH of fruit juices and storage temperature of fruits and juices may also favour growth of pathogenic organisms. Preparation of freshly squeezed fruit juices do not involve any step that could reduce the level of pathogens[15] in it, if already contaminated. Street foods are frequently associated with diarrhoeal diseases due to their improper handling and serving practices[10],[13],[15]. The fresh juices being dispensed in large numbers on a daily basis pose a health risk to the huge proportion of society consuming them.

II. Aims And Objectives

1)**.**To isolate and identify the aerobic microbial contaminants of street vended fresh fruit juices. 2).To calculate the Total Viable Count (TVC), Total Coliform Count (TCC), Total Staphylococcal Count, Total Yeast Count, Total Salmonella Count and Total Vibrio Count of the contaminating flora, if any. 3).To determine the prevalence of Candida albicans and Non-albicans Candida in the fresh fruit juices.

III. Materials And Methods

It was a prospective cross sectional study done at Department of Microbiology , RIMS, Ranchi between September, 2017 to August, 2018. A total of 50 juice samples were collected from 29 different locations, situated in and around Ranchi city. Out of 50, 8 were Mousami, 7 Orange, 7 Pomegranate, 7

Pineapple, 7 Grape, 7 Plain Sugarcane extract and 7 other Sugarcane extracts which were mixed with mint and lemon extracts.

1.Collection of samples – The juices were collected in a 50ml sterile wide mouthed universal container and packed in box containing ice packs.

2.Transportation - The box was then transported immediately under appropriate conditions to Department of Microbiology at RIMS, Ranchi and the processing was started within 1 hour of collection.

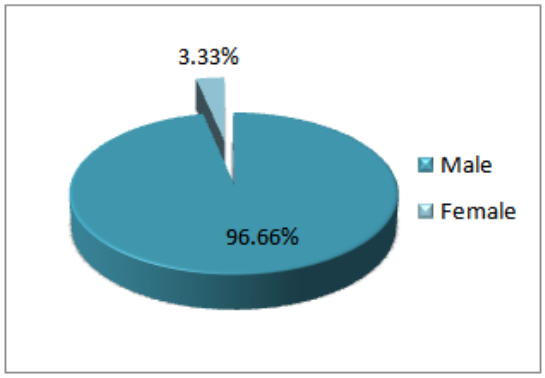
3.Procedure – In the Bacteriology section of our Laboratory, the box was opened in a designated clean area. Half of the juice sample was decanted into another sterile universal container. Sample in one container was used for pH measurement using digital pH meter while the remaining sample in the second container was used for inoculation onto the appropriate culture medias.

Using nichrome wire loop, which had the capacity to hold 0.001 ml of sample in it, 6 different media plates were inoculated. The plates inoculated were Mueller Hinton Agar(MHA), Mac Conkey Agar(MA), Xylose Lysine Deoxycholate Agar(XLD), Thiosulphate Citrate Bile Salt Sucrose Agar(TCBS), Mannitol Salt Agar(MSA) and Sabouraud's Dextrose Agar(SDA). One drop of sample (0.001ml) was placed in the centre of each plate and the drop was spread over whole surface area of the plate by spread plate technique. All the six plates were incubated aerobically for 24-48 hours at 37˚C.

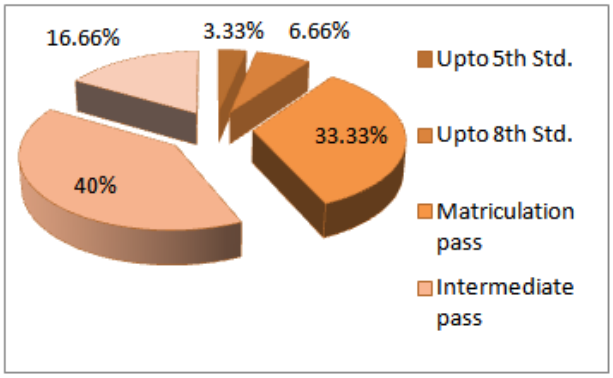
4.Identification -The organisms were identified by the help of their cultural characteristics, morphological features and biochemical properties. In case of any doubt, the help of B D Phoenix 100 was taken.

IV. Results

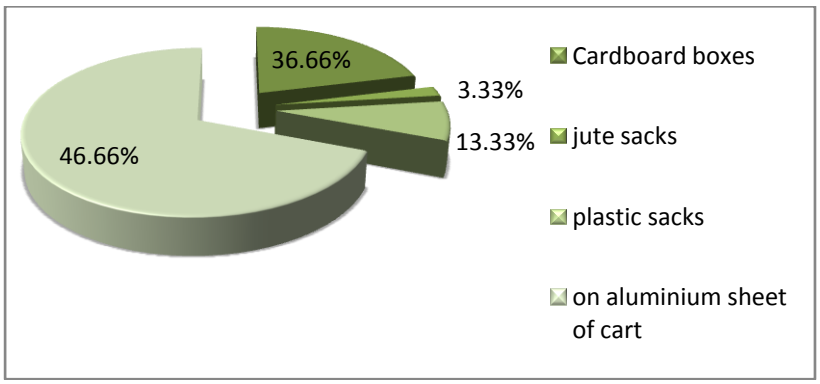
Out of 30 respondents, only one (3.33%) juice seller was female, while 29 (96.67%) were males. The female juice seller was educated only upto 5th standard (3.33%). Among the rest, 2 (6.66%) were educated upto 8th standard , 10 (33.33%) were matriculation pass, 12 (40%) had passes their intermediate exams and 5 (16.67%) were graduates. All (100%) the juice makers bought their fruits on a regular basis from the Daily market, Main road, Ranchi. Out of the 30 juice sellers, 11(36.66%) kept the fruits stored in cardboard boxes throughout the day. 1 (3.33%) juice seller kept them in jute sacks, 4(13.33%) in plastic sacks and 14 (46.66%) stored them on the aluminium sheet spread on their mobile cart.



Graph 1: Pie chart depicting the distribution of male and female juice sellers in the study.



Graph 2: Pie chart depicting standard of education among the juice sellers.

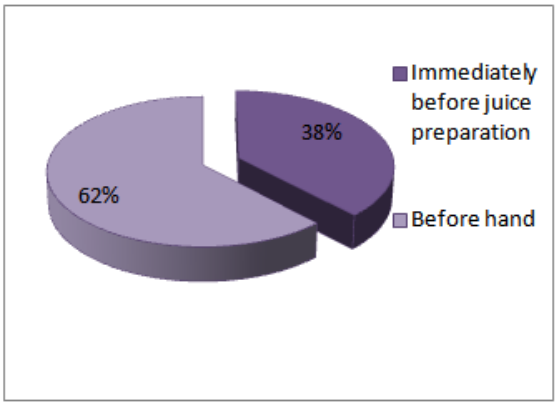


Graph 1: Pie chart depicting the ways adopted by juice sellers for storage of fruits.

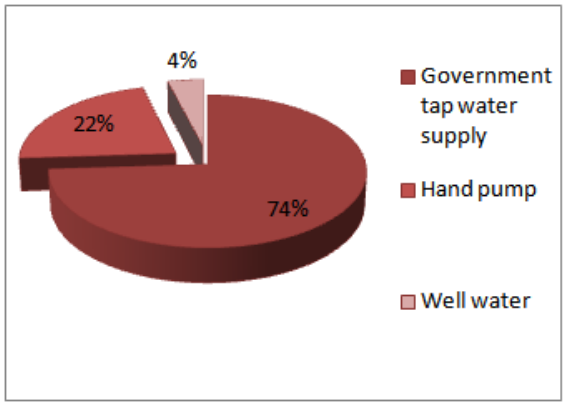
All the latter 14, were sugarcane juice sellers who had already peeled off the sugarcanes in the morning itself before starting to sell juices and stored them uncovered throughout the day on the juice vehicle exposed to all types of contaminants coming from dust and environment.

Out of 50 juice samples, 19(38%) of them were prepared from fruits that had been washed and peeled immediately before making the juice, while 31(62%)(this included all the 14 sugarcane juices) of them were made from fruits which had been already washed and peeled off before hand.

Out of the total 50 juice samples, 37 (74%) were prepared using tap water supplied by government to different localities within Ranchi. 11 (22%) of the juice sellers used water from Hand pump while a minority of them, 2 (4%) used well water for fulfilling different purposes like washing of the juicer machine, fruits, utensils, tumblers and for handwashing.



Graph 2: Pie chart depicting practices of juice sellers regarding washing and peeling of fruits.



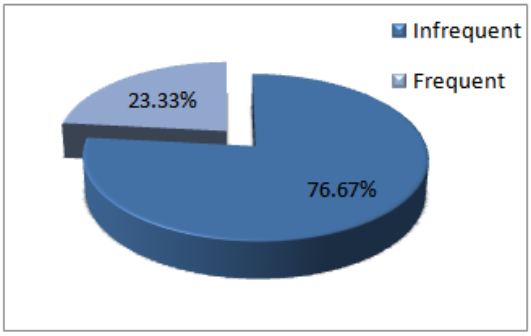
Graph 3: Pie chart depicting the use of water from various sources.

23 (76.67%) juice handlers were observed to practice infrequent handwashing, while rest 7 of them (23.33%) were into frequent handwashing.

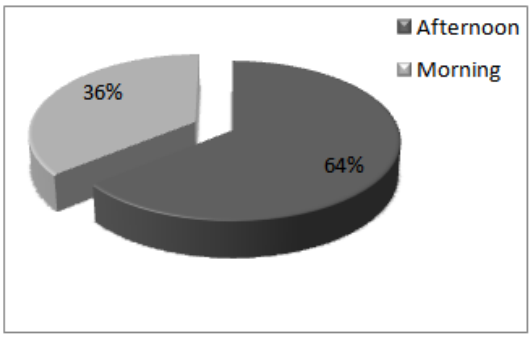
Frequent handwashing implied that the juice makers washed their hands before starting the preparation, using water. The others did not wash their hands before starting to prepare the juice. They only washed hands if they felt the need to do so.

Out of 50, 32 (64%) juices were collected in the afternoon and 18 (36%) in the morning (Before 12 noon). All the 30(100%) juice sellers were in the habit of receiving or handling currency, amidst juice preparation. They were not aware that even currency notes and coins can carry contaminant pathogens which can get transmitted through their hands into the juice while preparation.

14 (28%) juices collected had an alkaline pH while 36(72%) had acidic pH. All the juices having alkaline pH were sugarcane juices.



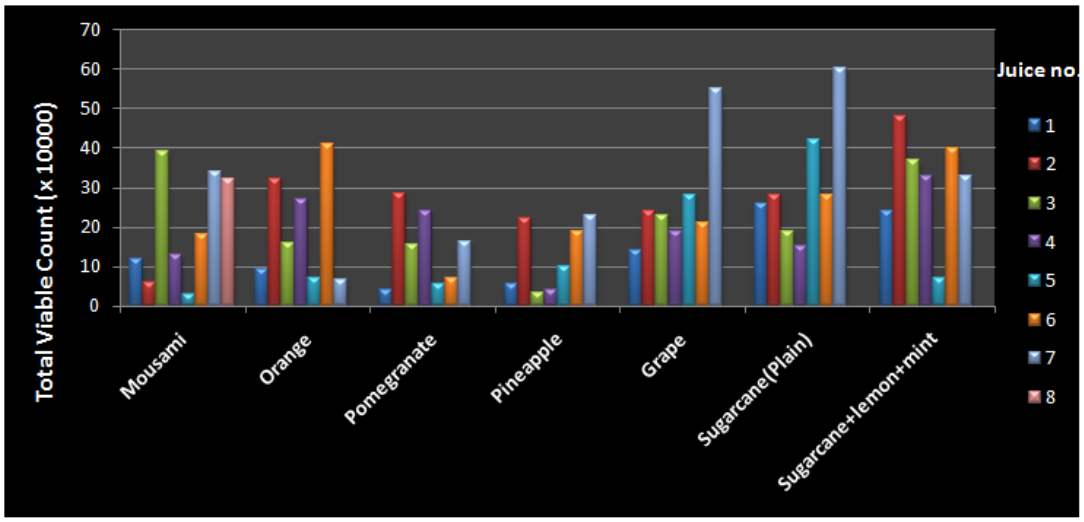
Graph 4: Pie chart depicting frequency of handwashing adopted by juice makers.



Graph 5: Pie chart depicting the time of collection of juice samples.

The organisms isolated were *Enterobacter aerogenes, E.cloacae, Klebsiella pneumonia, K.oxytoca, Pantoea agglomerans, Leclercia adecarboxylata, Lactobacillus acidophilus, L.casei, S.aureus, Pseudomonas* spp., *Proteus mirabilis, Candida* spp., *Streptococcus* spp. and *Micrococcus* spp.

The highest TVC was recorded in the plain sugarcane juice followed by grape juice, sugarcane juice mixed with mint and lemon extract, orange juice, mousami juice, pomegranate juice, pineapple juice.

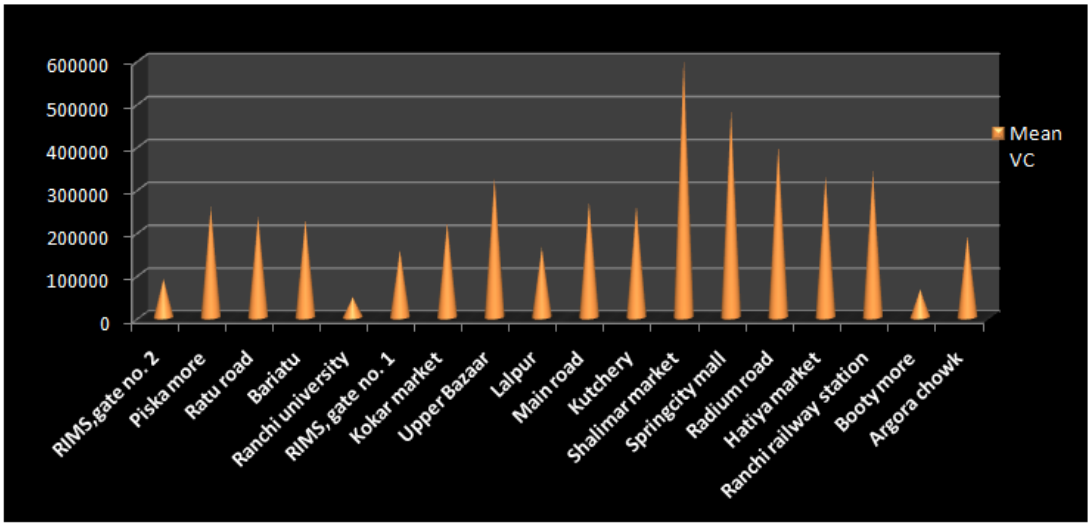


Graph 8 : Bargraph showing Total Viable Count in various juice samples.

The TVC for mousami juices ranged between 6×10^4 - 39×10^4 CFU/ml and the mean VC being 19.63×10^4 CFU/ml ; for orange juices ranged between 6.5×10^4 - 41×10^4 CFU/ml; for pomegranate juices ranged between 4×10^4 – 28.4×10^4 CFU/ml; for pineapple juices ranged between 3.5×10^4 - 23×10^4 CFU/ml; for grape juices ranged between 14×10^4 - 55×10^4 CFU/ml; for plain sugarcane juices ranged between 15×10^4 - 60×10^4 CFU/ml; for sugarcane juices mixed with mint and lemon extract ranged between 7×10^4 - 48×10^4 CFU/ml.

Mean VC for juices obtained from RIMS, Gate no. 2 was 9×10^4 CFU/ml, Piska more was 25.73×10^4 CFU/ml, Ratu road was 23.52×10^4 CFU/ml, Bariatu was 22.8×10^4 CFU/ml, Opposite Ranchi university was 4.73×10^4 CFU/ml, RIMS Gate no. 1 was 15.625×10^4 CFU/ml, Kokar market was 22×10^4 CFU/ml, Upper Bazaar was 32.5×10^4 CFU/ml, Lalpur was 16.33×10^4 CFU/ml, Main road was 26.85×10^4 CFU/ml, Kutchery was 26×10^4 CFU/ml, Shalimar market was 60×10^4 CFU/ml, Outside Springcity mall was 48×10^4 CFU/ml, Radium road was 40×10^4 CFU/ml, Hatiya market was 33×10^4 CFU/ml, Ranchi railway station was 34.1×10^4 CFU/ml, Booty more was 6.5×10^4 CFU/ml and for the juice obtained from Argora chowk was 19×10^4 CFU/ml.

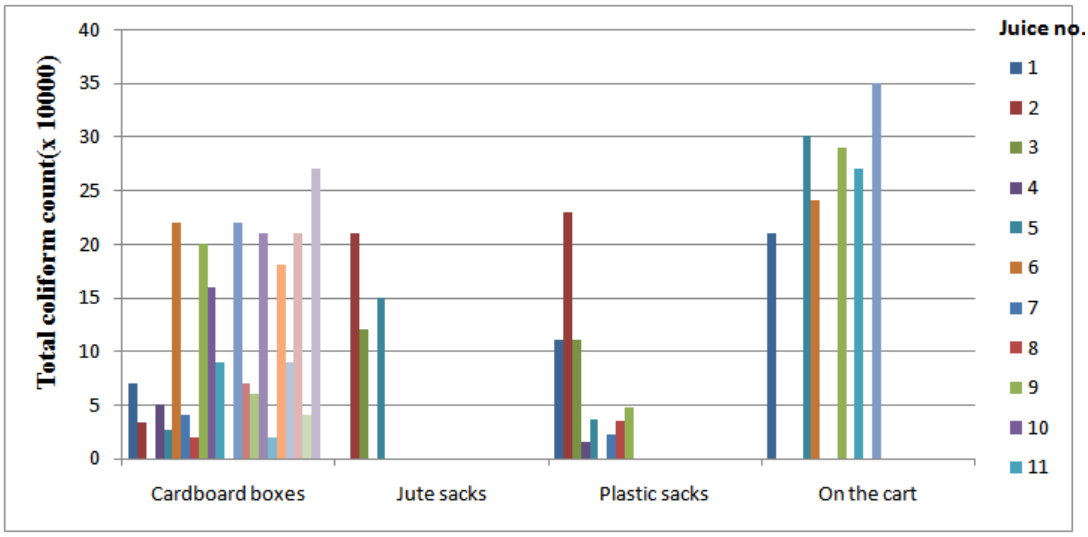
The highest mean Viable count was observed in juice obtained from Shalimar market, followed by the vendor outside spring city mall, Radium road, Outside Ranchi railway station, Hatiya market and Upper Bazaar. The lowest mean was recorded in juices obtained from Opposite Ranchi university.



Graph 9 : Bargraph showing Mean of Viable counts in juices obtained from different locations in Ranchi city.

The TCC in mousami juices ranged between 0×10^4 - 27×10^4 CFU/ ml and mean Coliform count being 11.875×10^4 CFU/ml; in orange juices ranged between 2×10^4 - 23×10^4 CFU/ ml; in pomegranate juices ranged between 0×10^4 - 11×10^4 CFU/ ml; in pineapple juices ranged between 1.5×10^4 - 21×10^4 CFU/ ml; in grape juices ranged between 5×10^4 - 22×10^4 CFU/ ml; in Sugarcane (plain) juices ranged between 0×10^4 - 30×10^4

CFU/ ml; in sugarcane juices mixed with mint and lemon extract ranged between 0×10^4 - 35×10^4 CFU/ ml and the mean Coliform count being 13×10^4 CFU/ml.



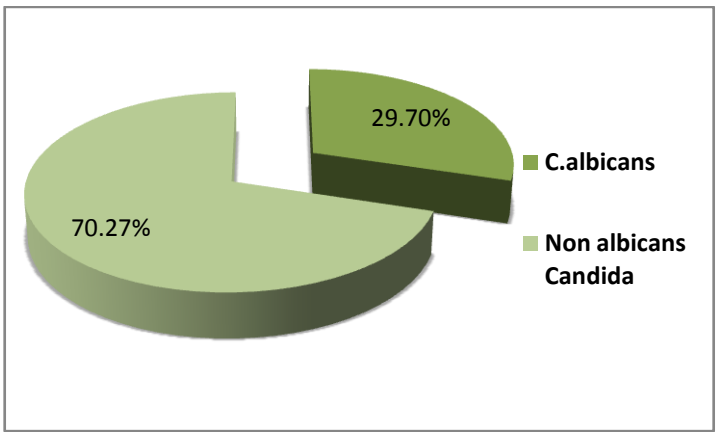
Graph 60: Bargraph showing Total Coliform Counts in various juices in relation to the place of temporary storage.

The highest TCC was recorded in sugarcane juice mixed with mint and lemon extract, followed by Plain Sugarcane, Mousami, Orange, Grape, Pineapple and Pomegranate.

The mean of Coliform counts was calculated and found to be highest in juices obtained from Radium road followed by Outside Springcity Mall, Upper Bazaar, Outside Ranchi railway station and Kutchery road. The lowest mean was for those obtained from Shalimar and Hatiya market.

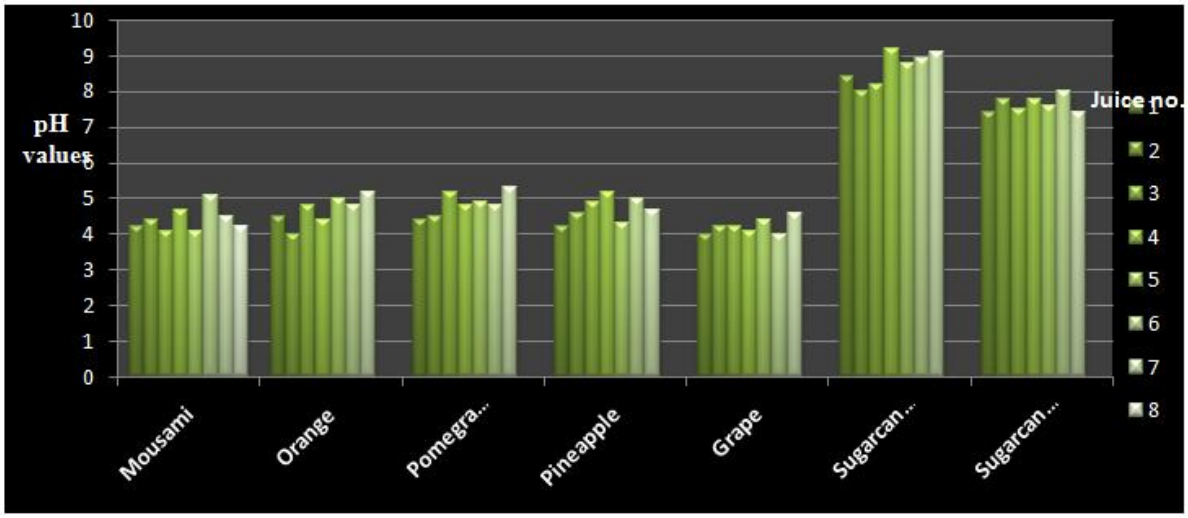
The highest YC was recorded in Grape juice, followed by Orange, Pomegranate, Mousami, Pineapple, Sugarcane juice mixed with mint and lemon extract and Plain sugarcane. The Highest mean value for Yeast counts was observed in juices collected from Main road followed by Argora chowk, Outside Ranchi railway station, Piska more, Ratu road, Bariatu area and RIMS gate no. 2. The lowest mean was from Kutchery road, Shalimar market, Outside Sprincity mall, Radium road and Hatiya market.

Out of 37 yeast isolates obtained by plating, 26 (70.27%) did not form Germ tube on incubation in pooled human serum at 37˚C for 2 hrs. Hence, GTT showed predominance of Non-albicans Candida species over Candida albicans 11 (29.7%).



Graph 11 : Pie chart depicting prevalence of Candida albicans and Non-albicans Candida in street vended fresh fruit juices.

Staphylococcus aureus was isolated only from Sugarcane juices (both plain and mixed with mint and lemon extract). The TSC in plain sugarcane juices ranged between 4×10^4 - 60×10^4 CFU/ml. Mean staphylococcal count in plain sugarcane juices was calculated to be 15.57×10^4 CFU/ml. The TSC in sugarcane juices mixed with mint and lemon extract ranged between 18×10^4 - 36×10^4 CFU/ml. Mean staphylococcal count calculated was 14.86×10^4 CFU/ml.



Graph12 : Bargraph depicting pH values of various juice samples.

V. Discussions

In this study, 29 out of 30 juice sellers were males and 1 was female while in studies conducted at Hyderabad[1], Guwahati[6] and Nairobi[14], all the respondents were male subjects. Similar to this study, the total respondents constituted of both male and female juice sellers in studies done at Nigeria[20], Ghana[17] and Malaysia[5]. 65% of the fruit juice makers were males and 35% were females in study done in Ogun State, South Western Nigeria[3]. In the present study, gender of the juice seller was not found to significantly affect the TVC (p= 0.833), TCC (p= 0.253), TYC (p= 0.842), TSC (p= 0.966) of the juices they prepared.

In this study, 40% and 43.33% of juice vendors had completed their secondary and primary education respectively, while in the study done at Hyderabad[1] 30% and 46% of them had completed their secondary and primary education respectively. Similar to my study, 43.33% of juice makers were educated upto secondary school in the study done at Bahirdar Town, Northwest Ethiopia[4]. In the present study, educational status of juice sellers was not found to significantly influence the TVC (p= 0.683), TCC (p=0.134), TYC (p=0.915), TSC(p=0.728) in the juices they prepared. Here, we infer that even the juices obtained from vendors who were enough educated, were equally contaminated with microbial flora as were the juices obtained from less educated vendors.

In this study as well as in other similar studies done in different cities of India, none of the juice makers used refrigerators for temporary storage of fruits while 10% of the juice sellers stored their fruits in refrigerator in the study done at Ogun State, South Western Nigeria[3] and 6.7% of them did the same in Bahirdar town, Northwest Ethiopia[4]. Hence, In our study, fruits were stored at room temperature which will be helpful in proliferation of certain microbes over the surface of fruits and these microbes will find their way into the juices through the improperly washed fruits. The place of storage was found to be statistically significant in the appearance of elevated TCC(p= 0.005), TYC (p= 0.000) and TSC (p= 0.020). The Total Coliform count was overall higher in cases of juices made from fruits which were peeled and stored uncovered on the aluminium spread of the mobile sugarcane juice cart and the reason can be attributed to the ambient temperature for proliferation of microbes, exposure to dusts and flies, time elapsed between peeling and juice preparation during which microbes can multiply a number of times.

In the present study, 62% of the juices collected were made from fruits which were peeled beforehand and stored in open and only 38% were immediately peeled and washed before juice preparation. On the contrary, 73.3% of the respondents peeled and cut the fruits at the time of juicing in the study conducted at Bahirdar Town, Ethiopia[4]. The time elapsed between peeling, washing of fruits and juice preparation was found to be statistically significant in my study, in affecting the TVC (p= 0.000), TCC(p= 0.000) and TYC (p= 0.013). This is probably because storing of peeled fruits will allow the contaminants to multiply on its surface until the fruit is used for juicing.

In our study, 76.67% of the juice sellers were in habit of infrequent handwashing and 23.33% washed their hands frequently while, on the contrary, in the study done by Lakshmi et al in Hyderabad[1], 78% practices handwashing with water before every juice preparation. In another study done by Asmamaw et al at Bahirdar Town, Ethiopia[4], 62.5% of the respondents washed their hands frequently (before juice preparation). In the present study, handwashing was not found to be statistically significant in determining the contamination in the juices. TVC (p= 0.119), TCC (p= 0.236), TYC (p= 0.340), TSC (p= 0.151).

In the present study the juices from crowded areas like market places, outside large Malls of the city, railway station etc. were found to be more contaminated. The association between location of juice vendors and TVC (p= 0.017), TCC (p= 0.009) and TSC (p= 0.000) was found to be statistically significant but association with TYC (p= 0.900) was not significant. Similar to the results in my study, Tambekar et al[7] in his study done at Amravati city also concluded that juices from crowded sites were more contaminated (55%) than that of the less crowded places (45%).

In this study, the juice type was found to be statistically significant in association with TCC(p= 0.006), TYC(p= 0.000), and TSC(p= 0.010). *Staphylococcus aureus* was isolated only from sugarcane juices, probably due to the alkaline pH of these juices. In the study done by Asmamaw et al[4], *S.aureus* was isolated from all the juice samples and they attributed its presence to use of dirty clothing and contaminated hands of food handlers, which indicated lack of knowledge of hygienic practices and food safety. In another study done by Bello et al[3], almost all the juices showed the presence of *S.aureus* in them which they thought to be due to contamination via handling. It is unlikely for the introduced *S.aureus* to survive in juices having low pH, it is possible that they may do so in juices having pH values more than 4 [11].

In the present study, all the other juices apart from sugarcane, were citric fruit juices having acidic pH and that is why those were found to be constantly associated with isolation of yeast species. The yeast isolation can be attributed to the sugar content in the juices and the acidic pH which is favourable for their survival.

In this study, pH of the juices were found to be statistically significant in terms of TVC(p= 0.025), TCC(p=0.001), TYC(p=0.000) and TSC(p=0.000).

In this study, all the juice samples tested were devoid of *Salmonella* spp., *Shigella* spp. and *Vibrio* spp. Similar to the present study, *Salmonella* spp. and *Shigella* spp. were not isolated in any juice samples tested by Durgesh et al[8] in his study conducted in Mumbai city but they found presence of *Vibrio cholerae* in 60% of sugarcane juice and 50% of carrot juice. Durgesh et al isolated *E.coli* in almost all the samples they tested while on the contrary, *E.coli* was not isolated from any juice sample tested in the present study.

VI. Conclusion

The demand for fruit juices has been increasing due to their health benefits which in turn, has led to increase in disease outbreaks linked mainly to fresh fruit juices in recent years. The knowledge on food safety measures was observed to be overall good among the juice sellers but the same was not practiced routinely by them. The peeling of fruits hours before and using wet towels to cover them for preventing the fruits from becoming dry, were some of the major issues which need to be intervened. Since the juices were found to be contaminated with pathogenic organisms, regular monitoring of the quality must be performed to avoid any outbreak of food borne illnesses.

References

[1]. S.G.D.N. Lakshmi Reddi, R.Naveen kumar, N.Balakrishna, V. Sudershan Rao. Microbiological quality of street vended fruit juices in Hyderabad, India and their association between food safety knowledge and practices of fruit juice vendors. Int. J. Curr. Microbiol. App. Sci (2015) 4 (1) : 970-982.

[2]. Kamal Rai Aneja, Romika Dhiman, Neeraj Kumar Aggarwal, Ashish Aneja. Emerging preservation techniques for controlling spoilage and pathogenic microorganisms in fruit juices.2014; Internal journal of Microbiology; Hindawi Publishing Corporation. Pg 1-14.

[3]. Bello OO, Bello TK, Bankole SA(2013). Occurrence of antibiotic resistant Staphylococcus aureus in some street vended foods in Ogun state, Nigeria. Journal of Advances in biology, 1(1):21-28.

[4]. Asmamaw Leul, Mulugeta Kibret. Bacteriological safety of freshly squeezed mango and pineapple juices served in juice houses of Bahirdar Town, Northwest Ethiopia. IJSBR; 2012; Volume 6, No. 1,Pg 24-35.

[5]. Mutalib Noor-Azira Abdul, Mohammad-Faid Abdul- Rashid, Shuhaimi Mustafa, Syafinaz Amin-Nordin, Rukman Awang Hamat, Malina Osman. 2012. Knowledge, attitude and practices regarding food hygiene and sanitation of food handlers in Kuala Pilah, Malaysia. Food Control. 27: 289-293.

[6]. Choudhury, M., Mahanta, L., Goswami, J., Mazumder, M. & Pegoo, B. 2010. Socioeconomic Profile and food safety knowledge and Practice of street food vendors in the city of Guwahati, Asaam, India. Food control. 22(2): 196-203.

[7]. Tambekar D.H., Murhekar S.M., Dhanorkar D.V., Gulhane P.B., Dudhane M.N., Quality and safety of street vended fruit juices : a case study of Amravati city , India. Journal of Applied Bioscience , 14, pp. 782-787,(2009).

[8]. Durgesh PM, Ranjana GK, Varsha KV. Microbiological analysis of street vended fruit juices from Mumbai City, India. International Journal Food Safety 2008; 10: 31-34.

[9]. Suaads A. , Eman A. H. Microbial growth and chemical analysis of bottled fruit juices and drinks in Riyadh , Saudi Arabia . Research journal of Microbiology , 3, pp.315-325, (2008).

[10]. Nicolas B, Razack BA, Yollande T, et al. street-vended foods improvement. Contamination mechanisms and application of food safety objective strategy: Critical review. Pak J Nutr 2007; 6: 1-10.

[11]. Lateef A, Oloke JK, Kana EB and Pachecho E.2006. The Microbiological quality of Ice used to cool drinks and foods in Ogbomoso Metropolis Southwest Nigeria. Internet journal of Food Safety. 8: 39- 43.

[12]. Lewis JE Thompson P , Rao C Kalavati , Rjanna B (2006) . Human bacteria in street vended fruit juices : A case study of Vishakhapatnam City , India . Internet J. Food Safety , 8 : 35-38.

[13]. Oliveira ACG, Seixas ASS, Sousa CP, Souza CWO. 2006. Microbiological evaluation of sugarcane juice sold at street stands and juice handling conditions in Sao Paulo, Brazil. Cad. Saude Publica, Rio de Janeiro. 22(5): 1111-1114.

[14]. Muinde OK, Kuria E. 2005. Hygienic and sanitary practices of vendors of street foods in Nairobi, Kenya. AJFAND online www.ajfand.net.5(1):1-13.
[15]. Victorian Government Department of Human Services, Food Safety Unit Melbourne, Victoria, 2005. Microbiological survey of freshly squeezed juices from retail businesses across Victoria. Available at: http://www.health.vic.gov.au/foodsafety (accessed September 9 2007).
[16]. Al-Jedah J.H., Robinson R.K., Nutritional value and microbiological safety of fresh fruit juices sold through retail outlets in Qatar. Pakistan Journal of Nutrition, 1, pp.79-81 , (2002).
[17]. Mensah P. Yeboah-Manu D. Owusu-Darkok, A. Ablordey. 2002. Street foods in Accra, Ghana: How safe are they? Bull WHO. 80:546-554.
[18]. WHO(2002). Food safety and food borne illness. Fact sheet ,237, p7.
[19]. Kurowska E.M. , Spence J.D. , Jordan J., Wetmore S., Freeman D.J., Piche L.A.,Serratore P., HDL Cholesterol raising effect of orange juices in subjects with hypercholesterolemia. American Journal of Clinical Nutrition ,72 ,pp. 1095-1100,(2000).
[20]. Umoh, V.J., & Odoba, M.B. 1999. Safety and quality evaluation of street foods sold in Zaria, Nigeria. Food Control. 10:9-14.
[21]. Harrigan WF. 1998. Laboratory methods in Food Microbiology. Academic Press London.
[22]. Splittstosser, D.F., 1979. Fruits and fruit products. In : Food and beverage mycology. Ed. Beuchat, LR. Avi Publishing Co. Inc, Westport, Connecticut.

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