

Seasonal Bacterial Profiling of Street Vended Panipuri Sold in Jabalpur City of Madhya Pradesh

¹Marwaha Prachi, ²Pathak Smita & ³Singh Alpana

¹Research Scholar, Govt. M. H. college of home science and Science for women, Jabalpur, M.P. (India)

²Professor, Govt. M. H. college of home science and Science for women, Jabalpur, M.P. (India)

³Professor, Department of Food Sci. and Tech. Jawaharlal Nehru Agriculture University, Jabalpur, M.P. (India)

ARTICLE DETAILS

Article History

Published Online: 09 June 2018

Keywords

Food Borne Diseases, Food Safety, Street Food, Street Food Vendors

*Corresponding Author

Email: marwahaprach[at]gmail.com

ABSTRACT

Background: India is a country where each city boasts its own unique street food and a large percentage of population relishes on these delicacies. The most popular street food in India is panipuri. **Aim:** The present study undertaken to assess the season-wise bacterial profile of masala pani and mater sold with panipuri in Jabalpur city of Madhya Pradesh, India.

Method: Total thirty-six samples of panipuri were aseptically collected and analysed within one hour of procurement. Isolation, enumeration and identification of the bacteria were carried out following the standard procedure. **Findings:** Result shows that the total viable count of bacteria in masala pani during summer, rainy and winter season varied between 58.6-121.3×10⁵CFU, 97-183×10⁵CFU & 30.6-66.6×10⁵CFU and mater is varied between 48-119.3×10⁵CFU, 83.6-130×10⁵CFU & 19.6- 45.6×10⁵CFU respectively. This finding shows that the bacterial contamination in panipuri samples were higher in rainy season followed by summer and winter season. Almost all panipuri samples have high microbial load of pathogenic bacteria such as *Enterobacter faecalis*, *Salmonella sp.*, *Pseudomonas fluorescense*, *Protease sp.*, *Streptococcus sp.*, *E. coli*, *Enterobacter aerogenes* & *Klebsiella pneumoniae*. The mean scores of total viable count (CFU) of masala pani and mater sold in Jabalpur city during different season do differ significantly ($p \leq 0.05$). Thus, bacterial profile demonstrates that the panipuri sold in Jabalpur city constitute an important potential hazard to human health. The emerging needs like provision of health education to the vendors and enforcing implementation of appropriate hygienic practice should follow the control measure to improve the microbial quality of panipuri.

1. Introduction

India is famous for their unique street food and millions of people consume a wide variety of ready to eat street foods and beverage. In developing countries like India street food contributes to 40 % of daily diet of urban population (consumer international, 2011). People who depends on these types of food are often more interested in its convenience rather than its safety, quality and hygiene.

Consumption of this type of foods potentially increases the risk of food borne diseases caused by various pathogens. Usually vendors sold these foods by wheels barrows, trays mats, tables and make shift stalls consequently, they increasing the risk of food contamination (Ray and Mishra 2014). Contamination also from raw material and equipment's, additional processing conditions, improper handling and prevalence of unhygienic conditions contribute significantly to the entry of food borne pathogens. The potential for the contamination of street food with pathogenic microorganism has been well documented and several disease outbreaks have been traced to consumption of contaminated street foods. (Abdussalam and Kaferstein, 1993). Microbial contamination of street food is an indicator of poor sanitary practices in the preparation and storage of the food. (Saxena and Agrawal, 2013).

In India street food hawkers are commonly unaware of food regulations and untrained in food related matters as well as disease outbreaks as they are from rural background and most of them are illiterate. Today, street food has become one of the major concern of public health and a focus for governments and scientists to raise public awareness. (Sharma and Mazumdar, 2014). Among all type of street foods, panipuri or golguppa is traditional and very popular in all cities of India. Therefore, taking these factors into account this study was undertaken to assessed the bacteriological quality of pani puri, which lavishly consumed at street sides in Jabalpur city, MP, India.

2. Materials and Methods

2.1. Materials

Nutrient Agar, various selective and differentiated media and IMVic Test kit were procured from Hi-Media, Mumbai and prepared as per Manufacture's instruction.

2.2. Sample site and Sample collection:

Microbiological investigation of pani puri sold in Jabalpur city were performed during the summer, rainy and winter season in 2016-2017. Each sample of panipuri was fragmented into two different segments (the liquid masala pani and solid mater masala) and were collected in sterile plastic containers, which

were sealed and transported to the laboratory and processed within 1 hour of collection.

2.3. Microbiological Analysis:

For the microbiological analysis of food samples, dilution was made according to the method given by Verma and Verma, (2016) After serial dilution pour plate technique was applied on nutrient agar for bacteria and potato dextrose agar for fungi. After solidifying, Petri plates were incubated at 37°C for 24 hours for bacteria and 28°C for 7 days for fungi in inverted position in B.O.D incubator. After completing the incubation period, count the colonies appeared on the surface of NA and PDA media in the Petri plates. This number is designated as colony forming unit (CFU) and it can be calculated by using the following formula (Verma and Verma, 2016).

$$CFU/g = \frac{\text{Number of colonies}}{\text{Weight of sample}} \times \text{Dilution factor}$$

The isolated colonies of organism were transferred into nutrient agar (for bacteria) and potato dextrose agar (for fungi) slant for maintenance and further identification.

2.4. Identification and isolation of Bacteria:

Gram staining, growth on selective & differential media and Biochemical tests for various bacterial isolates were done for identification of bacteria. After identification, pure culture stored in the culture collection centre in laboratory for further use and maintenance.

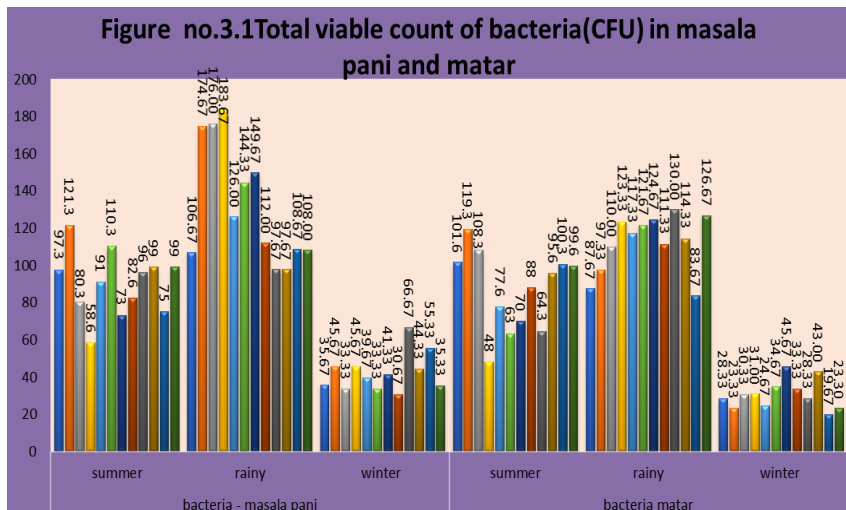
2.5. Statistical analysis

Statistical analysis was done using statistical package for the social sciences (SPSS) 16.0 version. All the values were expressed as Mean, SD and One-way ANOVA. Statistical significance level was considered to be present when the two-tailed probability was less than 0.05.

3. Result

A total of thirty-six panipuri samples in different seasons were analysed for presence of microbial pathogens by pour plate technique at different dilutions.

The figure no. 3.1 shows the total viable count of bacteria in masala pani is varied between 58.6-121.3x10⁵ CFU, 97-183x10⁵ CFU & 30.6-66.6x10⁵ CFU and mater is varied between 48-119.3x10⁵ CFU, 83.6-130x10⁵ CFU & 19.6-45.6x10⁵ CFU during the summer, rainy and winter season respectively.



The figure no. 3.1 shows the total viable count of bacteria in masala pani is varied between 58.6-121.3x10⁵ CFU, 97-183x10⁵ CFU & 30.6-66.6x10⁵ CFU and mater is varied

between 48-119.3x10⁵ CFU, 83.6-130x10⁵ CFU & 19.6-45.6x10⁵ CFU during the summer, rainy and winter season respectively.

Table no.3.1: Season-wise comparison of bacteria (CFU) in panipuri (masala-pani & matar)

Bacteria – Masala Pani			Bacteria -Matar		
Summer	Rainy	Winter	Summer	Rainy	Winter
97.3±2.5	106.7±0.6	35.7±3.2	101.6±4.7	87.7±1.5	28.3±2.1
121.3±5.1	174.7±3.1	45.7±2.1	119.3±1.52	97.3±1.2	23.3±1.5
80.3±1.5	176±1	33.3±2.1	108.3±3	110±2	30.3±0.6
58.6±2.5	183.7±3.1	45.7±3.2	48±1.5	123±2.5	31±1.7

91±2	126±3	39.7±1.5	77.6±2**	117.3±1.2	24.7±0.6
110.3±2.5	144.3±4.5	33.3±2.1	63±4.5	1.5±121.7	34.7±1.5
73±3	149.7±4.5	41.3±2.1	70±2	124.7±2.5	45.7±2.1
82.6±1.5	112±3	30.7±3.1	88±2	111.3±2.1	33.3±1.2
96±2.6	97.7±3.2	66.7±3.1	64.3±5	130±1.7	28.3±1.5
99±2	97.7±2.9	44.3±2.1	95.6±1.5	114.3±27.5	43±2
75±1	108.7±2.5	55.3±3.5	100.3±2.5	83.7±28	19.7±1.5
99±2	108±2	35.3±3.2	99.6±3	126.7±0.6	23.3±1.5

* significant difference at 0.05%

Table no.3.1 shows the season wise comparison of bacterial total viable count of masala pani and mater. The result shows that the mean scores of total viable count (CFU) of masala pani and matar in different seasons in Jabalpur differ significantly ($p \leq 0.05$). The bacterial contamination in panipuri samples were higher in rainy season followed by summer and winter season. Tambekar et al. (2008) and Bagumire and Karumuna (2017) also found in their studies that the microbial contamination is higher in rainy season compare to summer and winter season.

The majority of samples were found to contain the presence of different species of pathogenic microorganisms indicating poor microbiological quality of the food samples. Isolation and

identification of bacteria was done by streaking selected colonies on different selective media plates like Eosin Methylene Blue Agar (EMB), MacConkey Agar, Cetrimide Agar, Salmonella Shigella Agar and Blood Agar. Table no.2 shows the gram staining, morphological structure of identified bacterial isolates. Table no 3 shows the result of biochemical test of pure culture isolates. Based on the growth on selective and differential media and biochemical tests, various bacterial isolates were identified as (figure no.3) Enterobacter faecalis, Salmonella sp., Pseudomonas fluoresces, Protease sp., Streptococcus sp., E. coli, Enterobacter aerogenes and Klebsiella pneumoniae. Saxena et al. (2016) & Gulati and Chakraborty (2017) are also found similar results in their studies.

Table 2: Gram staining, morphological structure and identified bacteria from panipuri (Pani and Mater)

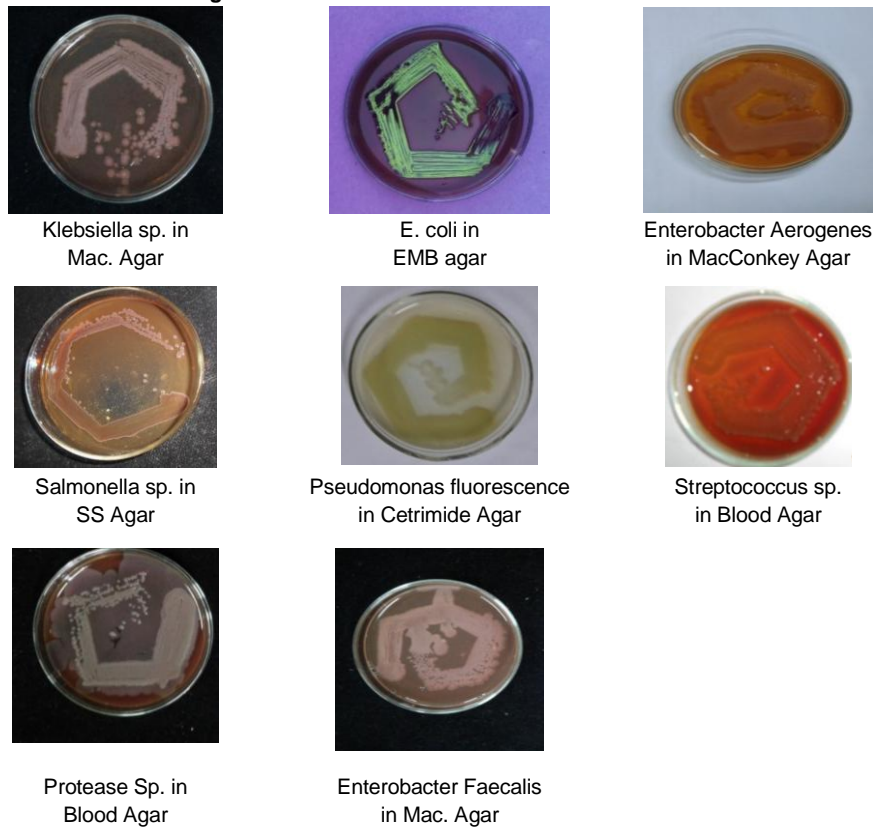
Food sample	Gram staining	Morphological structure	Identified bacteria
Pani puri – masala pani	-	Rod chain	Enterobacter faecalis
	-	Rod	Salmonella sp.
	-	Rod chain	Pseudomonas fluorescence
	-	Rod chain	Protease sp.
	+	Coccus	Streptococcus sp.
	-	Rod	E. coli
Mater	-	Rod	Enterobacter aerogenes
	-	Rod	E. coli
	-	Rod	Klebsiella pneumonia
	-	Rod	Salmonella sp.

Table no 4 3: Biochemical Test Results of Identified Bacteria

	gm	ind	mr	vp	Cat	glu	suc	cit	mot	amy	ure	cas	Oxi
E coli	-	+	+	-	+	+	+	-	+	-	-	-	-
Salmonella sp.	-	-	+	-	+	+	-	-	+	-	-	-	-
Klebsiella sp.	-	-	-	-	+	+	+	+	-	+	+	-	-
Pseudomonas fluoresces	-	-	-	-	+	-	-	+	+	+	-	+	-
Protease sp.	-	-	+	-	+	+	+	V	+	+	+	+	-
Streptococcus sp.	+	-	+	V	+	+	+	-	-	-	+	+	-
Enterobacter aerogenes	-	-	+	-	+	+	+	+	+	-	-	-	-

Enterobacter faecalis	-	-	-	+	+	+	+	+	+	-	V	-	-
Note: gm- gram stain, Ind-indole, Mr-methyl red, vp- Voges-Proskauer, Cat-Catalase, glu-glucose, suc- sucrose, cit- citrate, mot-motility, amy-amylase, ure-urease, cas- casinase, Oxi-Oxidase, V-11-89% positive													

Figure No 4.3: Master Plates of Bacterial Isolates



4. Discussion

Microbial contamination may be linked to factors such as equipment and utensils with inadequate hygienic condition, uncovered utensils and garbage bin. irregularity of hand washing, inappropriate processing, incomplete heating, use of contaminated water during preparation and washing or secondary contamination via contact with contaminated equipment's such as chopping boards, knives and serving wares (Derbew *et al.*,2013; Mahale *et al.*, 2008; Wai Q *et al.*,2006). This might also implicate the processing and rinsing water as possible sources of contamination of panipuri sold by street vendors (Das *et al.*,2010). The bacterial contamination in panipuri is because of the conditions under which it is prepared and vended. In most of the cases running water is not available at vending sites and thus hand and dish washing are usually done in buckets and sometimes without soaps. (Das *et al.*, 2012).

The occurrence of *E. coli*, *Enterobacter sp.*, *Klebsiella pneumonia* and *p. aeruginosa* may be due to poor personal hygiene of vendors, unhygienic handling of foods, poorly cleaned dishes and use of raw vegetables like onion etc. contaminated water supplies, vehicular transmission and sewage. (Tamberkar *et al.*, 2011). The detection of respiratory pathogens such as *Klebsiella sp.* in panipuri attributed to the bacterial aerosols generated due to sneezing and coughing in public places. (Das *et al.*,2010). Das *et al.* (2010) showed that

street food such as panipuri, bhelpuri and chaat in Bangalore city, were contaminated with high loads of pathogens that is *Streptococcus faecalis*, *E. Coli*, *Staphylococcus aureus*, *Bacillus sp.* *Klebsiella sp.* and *pseudomonas sp.*, which similar with our findings.

Hence, it is concluded that the bacterial contamination in panipuri samples were higher in rainy season followed by summer and winter season. panipuri was contaminated with bacteria like *Enterobacter faecalis*, *Salmonella sp.*, *Pseudomonas fluorescence*, *Protease sp.*, *Streptococcus sp.*, *E. coli*, *Enterobacter aerogenes* and *Klebsiella pneumoniae* which can cause various food borne diseases.

5. Conclusion

The present study shows high level of contamination in different season in one of the most popular Indian street food, panipuri sold in Jabalpur city. The study throws a light on the potential risk of food poisoning, to overcome this alarming situation, vendors should be taught to good manufacturing practice and good hygiene practice to reduce street foods contamination. Regular monitoring of the conditions of street vendors is necessary. The local government and the ministry should consider establishment of adequate facility and utility service as well as provision of necessary information, education and training programmes for vendors and consumers.

References

1. Alekhya Sabbithi, S.G.D.N. Lakshmi Reddi and R. Naveen Kumar. (2017). Identifying critical risk practices among street food handlers. *British Food Journal*, 119(2), 390-400. doi:10.1108/BFJ-04-2016-0174.
2. Alexopolous CJ, Mims CW and Blackwell M (1996) Introductory mycology (4th Ed), John Wiley and Sons publishers, Newyork, U.S.A. 868.
3. Arijit Das, G.S. Nagananda, Sourav Bhattacharya and Shilpi Bhardwaj. (2010). Microbiology Quality of Street - Vended Chaats Sold in Bangalore. *Journal of Biological Sciences*, 10(3), 255-260.
4. Barnett HL and Hunter BB (1972) Illustrated genera of imperfect fungi . 3rd edition, Burgess Publishing Co., 273
5. Booth C (1971) Genus *Fusarium*. Commonwealth Mycological Institute (CMI), Kew, Surrey, England. 273.
6. Booth C (1977) *Fusarium*. Commonwealth Mycological Institute, Kew, Surrey, England. 58pp.
7. Consumer International. (2011). "Serving up street food in the global south". available at: <http://streetfood.org> (accessed April 27, 2014),..
8. Deepti Gulati and Devirupa Chakraborty. (2017). Antibiogram of Bacterial Pathogens Isolated From One of The Most Popular Street Food (Panipuri) of Dehradun. *International Journal of Pharma and Bio Sciences*, 8(2), 395-400. doi:<http://dx.doi.org/10.22376/ijpbs.2017.8.2.b395-400>
9. Durgesh P. Mahale, Ranjana G. Khade and Varsha K. Vaidya. (2008). Microbiological Analysis of Street Vended Fruit Juices from Mumbai City, India. *Internet Journal of Food Safety*, 10 , 31-34.
10. Ellis MB (1971) Dematiaceous Hyphomycetes. CMI, Kew, Surrey, England. 608
11. Ellis MB (1976) More dematiaceous hyphomycetes. Kew: Commonwealth Mycological Institute. 507
12. FAO. (2007). "Spotlight: School Children, Street Food and Micronutrient Deficiencies in Tanzania",. FAO.
13. Gargi Saxena, Mukta Agrawal and Kailash Agrawa. (2016, october). Physico-chemical and microbiological quality assessment of street foods sold in Jaipur city of Rajasthan. *Food Science Research Journal*, 7(2), 270-275 . doi:DOI : 10.15740/HAS/FSRJ/7.2/270-275
14. Getu Derbew, Samuel Sahle and Mengistu Endris. (2013). Bacteriological Assessment of Some Street Vended Foods in Gondar, Ethiopia. *Internet Journal of Food Safety*, 15, 33-38.
15. Gilman JC (1957) A manual of soil Fungi Revised 2nd edition Oxford and IBH publishing Co. 220.
16. I Sharma and JA Mazumdar. (2014). Assessment of bacteriological quality of ready to eat food vended in streets of Silchar city, Assam, India. *Indian Journal of Medical Microbiology*, 32(2), 169--171.
17. Madhuchhanda Das & Chandni C. Rath & U. B. Mohapatra. (2012). Bacteriology of a most popular street food (Panipuri) and inhibitory effect of essential oils on bacterial growth. *Journal of Food Science and Technology*, 49(5), 564-571. doi:10.1007/s13197-010-0202-2
18. Mahendra Pal and Vijay J. Jadhav. (2013, December). Microbial Contamination of various Indian Milk Products. *Beverage & Food World*, 40(12), 43-44.
19. Manjit Kumar Ray and Piyush Kumar Mishra. (2014). A Preliminary Study of Fungi on Panipuri Sold in Different Areas of Dhubri Town, Assam. *Trends in Biosciences*, 7(11), 991-993.
20. Nagmani A, Kunwar IK and Manoharachary C (2006) Hand book of soil fungi: In International Pvt. Ltd. New Delhi, 477
21. Que-King Wei, Shu-Ling Hwang and Tong-Rong Chen. (2006). Microbiological Quality of Ready-to-eat Food Products in Southern Taiwan. *Journal of Food and Drug Analysis*, 14(1), 68-73.
22. Raper K and Fennel DI (1965) The genus *Aspergillus*. Williams and Wilkins, Baltimore, USA, 686.
23. Raper KB and Thom C (1949) A manual of the *Penicillia*. Williams and Wilkins Co., Baltimore, Md 920.
24. Saxena. G and Agarwal. M. (2013). Microbial Quality Assessment Of Street-Vended Gol Gappa And Bhelpuri Sold In Jaipur City of Rajasthan. *International Journal Of Food And Nutritional Sciences*, 2(1).
25. Subramanian CV (1971) Hyphomycetes An account of Indian species except *Cercosporae*. ICAR, New Delhi, 930pp.
26. Tambekar D H, Kulkarni R V, S D Shirsat and D G Bhadange. (2011, july). Bacteriological Quality of Street Vended Food Panipuri: A Case Study of Amravati City (Ms) India. *Bioscience Discovery*, 2 (3), 350-354.
27. Verma RK, Sharma N, Soni KK and Jamaluddin (2008) Forest fungi of central India. International Book Distributing Co. Lucknow, 418.