Section: Community Medicine

Original Research Article

A CROSS-SECTIONAL STUDY TO DETERMINE BACTERIOLOGICAL CONTAMINATION AND ITS RISK FACTORS AMONG PANIPURI VENDORS IN KALABURAGI CITY

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Abstract

Background: In most of the developing countries, food safety is becoming a serious threat to public health. In India, food borne diseases are expected to rise from 100 million in 2011 to 177 million in 2030. Nearly 1,20,000 deaths are reporting each year imposing a burden of over 8 million Disability Adjusted Life Years (DALYs). Street vended foods like panipuri are sold almost every corner of Kalaburagi and are popular among all the ages. **Aims/Objectives:** To determine the level of bacterial contamination in panipuri sold in Kalaburagi city and its associated risk factors. **Materials and Methods:** A cross-sectional study was conducted, with equal number of panipuri vendors selected from each of the four zones of the Kalaburagi city, within 10 km radius starting from Gulbarga Institute of Medical Sciences (GIMS) as centre of the city. Total 160 samples of panipuri (80 each of masala pani and puri, 20 from each of four zones) were aseptically collected in sterile containers and were processed at GIMS Microbiology Laboratory using standard microbiological techniques. **Results:** Among the 80 vendors participated in the study, 84% of them were contaminated with bacterial organisms in either masala pani or puri. Years of experience (OR = 8.145, 95% CI: 2.30-31.73), dressing of vendors (OR =6.698, 95% CI: 0.820-54.71) and personal hygiene (OR = 10.4, 95% CI: 1.71-201.1) were having significant associations with bacterial contamination, as revealed by binary logistic regression. **Conclusion:** This study demonstrates that bacterial contamination among pani puri vendors are due to lack of hand hygiene knowledge, infrequent cleaning of utensils and lack of proper waste management and there is a need of education and training for food handling techniques.

INTRODUCTION

As per Food and Agriculture Organization (FAO 1997) guidelines, street foods are defined as "ready-to-eat foods and beverages prepared and/or sold by vendors and hawkers, especially in streets and other similar public places."[1] These street-vended foods are mostly prepared on the street and ready to eat or prepared at home and served on the streets without further preparation.[2] India is a country where each city hosts its own culture and variety of dishes, including street foods. Among the street vended foods most popular is pani puri or golgappas. Pani puri also known as golgappa, puchka, or pakodi, culminates as the king of evening snacks. This street snack consists of three separate articles, i.e., pani, puri and masala. The pani is sour water to which spices like salt, black pepper, dry mango powder, cumin, mint leaves, etc. are added. Puri is a fried product made with refined wheat flour and masala is prepared with boiled or mashed potatoes and chickpeas mixed with spices. In every puri, masala is added after making a hole with a bare finger in it and then spicy water is filled into it and served to

the consumers in either disposable containers or plates on sites.[3-4] Foodborne disease incidence in India is anticipated to progress from 100 million in 2011 to 177 million by 2030. Each year, nearly 1,20,000 deaths are reported, inflicting a burden of over 8 million Disability Adjusted Life Years (DALYs). This means that by 2030, one out of every nine people will become ill.[5-6] Children under the age of five are the most vulnerable to foodborne diseases and at 40% more risk, with around 30,000 deaths each year.[6] By 2030, one in three members of wealthy urban households may suffer from a foodborne illness due to the ongoing processes of urbanization and GDP growth.[5] The Southeast Asian Region is prone to foodborne events due to its climatic conditions, deep-rooted food habits, poverty, inadequate basic hygienic and sanitary facilities, and low public awareness of food safety.[5,7-8] Street-vended foods like panipuri are sold almost in every corner of Kalaburagi and are popular among all ages. They are consumed by a huge population and frequently associated with food-borne illnesses due to their improper handling and serving. In light of this background, this study aims to find the level of bacterial contamination among panipuri vendors in Kalaburagi city, Karnataka.

Objectives: To determine the level of bacterial contamination in panipuri sold in Kalaburagi city and its associated risk factors.

MATERIALS AND METHODS

Study Design and Area

Kalaburagi is the largest district in the Kalyana-Karnataka region. A cross-sectional study was carried out in the major streets and markets of Kalaburagi City, Karnataka, from May 2023 to July 2023. This study was conducted with collaboration between the departments of community medicine and microbiology at the Gulbarga Institute of Medical Sciences
(GIMS), Kalaburagi. Panipuri vendors located within the 10-kilometer radius surrounding the Gulbarga Institute of Medical Sciences campus were considered for the study. Those who had not given informed consent were excluded from the study. The entire study was conducted in two steps. The first step involved mapping of Kalaburagi City. In the second step the entire city was divided into four zones with the help of major streets passing through the centre of the city and considering the GIMS campus as the centre point. [Figure 1]

Sample Size Calculation

This is an exploratory study done among pani puri vendors. According to the study done in 2019 by Mehta et al., 82% of the pani puri samples were contaminated.[9] Taking this as prevalence with 6 % precision, our sample size was around 160. Thus, a total of 160 samples (80 each of masala pani and puri) were collected from the selected vendors in Kalaburagi city. Considering the migratory pattern of vendors along with our limited time and resources, we purposively selected 80 vendors from the four zones of Kalaburagi city.

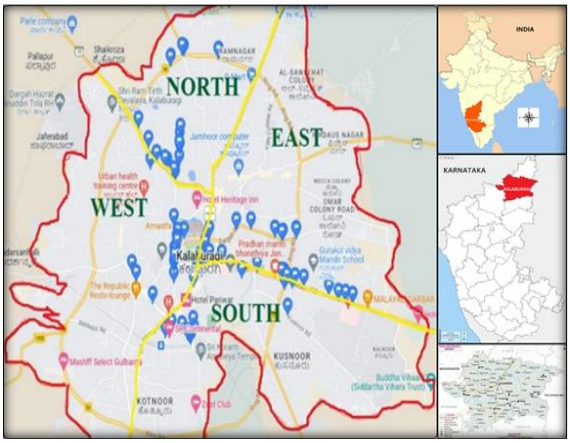

Figure 1: Map showing Kalaburagi city and the location of vendors in four different zones (courtesy: www.mapsofindia.com)

Figure 2: Working environment of panipuri vendor

We trained two field assistants to help the investigator with data collection. The data was collected based on face-to-face interviews after obtaining informed consent from the participants. A predesigned, semi-structured questionnaire was developed and pretested for clarity and validity. The four-page questionnaire consists of four sections with 40 variables: (i) socio-demographic characteristics of vendors; (ii) mode of preparation (source of water, preparation of puri, duration of keeping puri before utilization); (iii) observation of vendors, preparation site, and surroundings (site of shop, presence of drainage, dressing pattern of vendors, use of gloves, aprons during food preparation, use of hair caps, condition of nails, habit of keeping food containers closed); and (iv) food handling practices (frequency of washing

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utensils, washing hands between each preparation, cleaning the preparation area, types of plates used for serving). Prior to the interview, the vendors were assured of total confidentiality. [Figure 2]

Sample Collection

Two samples from each vendor containing masala pani and puri were collected in separate sterile screw–capped containers and transported aseptically to the microbiology laboratory of GIMS, Kalaburagi, for further processing within one hour. Puri will be broken and filled in a sterile container by the vendor itself. These samples were placed in an expanded polystyrene-moulded box with ice packs until they reached the laboratory. The samples were stored at a temperature of 2-4oC until sample processing started at the laboratory.

Isolation of bacteria

Each masala pani and puri item was homogenized prior to inoculation onto culture media. For this, a 1 g sample was mixed with 9 mL of sterile water, crushed, and grounded in a sterile mortar and pestle. Whereas, a loopful of pani samples was directly inoculated onto culture media without homogenization. Various culture media like nutrient agar, MacConkey agar, thiosulphate citrate bile-salt sucrose agar, xylose lysine deoxycholate agar, mannitol salt agar, and blood agar were used for the inoculation of samples, and the plates were incubated at 37°C for 24 hr. Turbidity in the broth indicated the presence of microbial growth, which was subsequently sub cultured on Cysteine Lactose Electrolyte Deficient Agar (CLED) and incubated for 24 hours at 37 ˚C (Figure 3).[10]

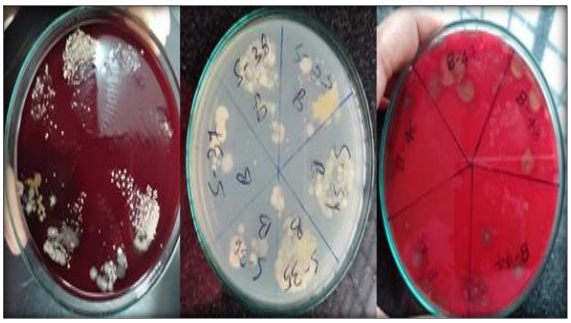

Figure 3. Sample processing and isolation of organisms

Identification of bacterial isolates

All the isolates were first differentiated by colony morphology and Gram staining reaction. They were further identified by an array of biochemical tests. We performed various biochemical tests such as oxidase, catalase, coagulase, DNAses, oxidative/fermentative, methyl-red, and Voges-Proskauer for gram-positive isolates. For Gram-negative isolates, indole, methyl red, Voges-Proskauer, citrate, triple sugar iron agar, catalase, oxidase, urease, and oxidative/fermentative tests were performed.[10]

Statistical Analysis

Statistical analysis was performed in SPSS version 16. The results were analyzed using descriptive statistics like frequency and percentage. A chi-square test was applied to test proportions. P value of <0.05 was considered statistically significant.

RESULTS

A total of 160 samples of masala pani and puri collected from different zones were analysed for the isolation of bacterial pathogens. Among the total samples collected, 96 (60%) were found to be contaminated with high loads of pathogenic bacteria. When we look at overall contamination, either the masala pani or puri in 67 (84%) vendors were contaminated. [Figure 4]

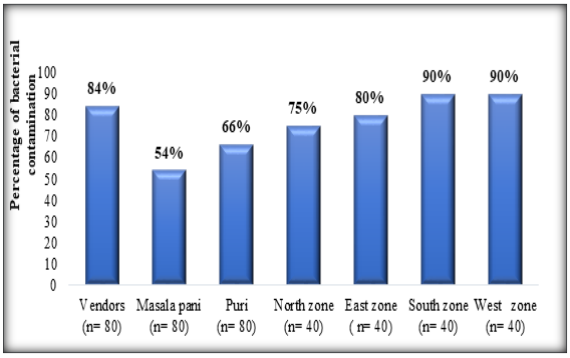

Figure 4. Percentage of bacterial contamination among vendors, collected samples, and zones of Kalaburagi

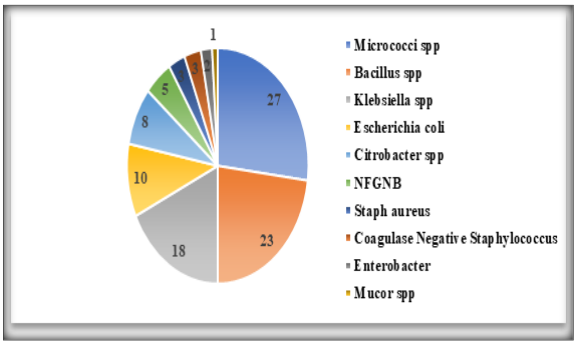

Figure 5. Percentage of microbiological organisms leading to contamination

The majority of samples were contaminated with Micrococci spp. (27.1%), Bacillus spp. (22.9%), Klebsiella spp. (17.7%), Escherichia coli (9.6%), Citrobacter spp. (8.3%), Non-fermenting Gram-negative bacteria (NFGNB) (5.2%), Staphylococcus aureus (3%), Coagulase negative staphylococcus (3%), Enterobacter spp. (2.1%), and Mucor spp. (1%). [Figure 5]

Demographic and job profiles of vendors

The median age of vendors was 30 years. Those vendors with age greater than or equal to 30 years, illiterate and experience less than 10 years were found to be significantly associated with panipuri contamination. [Table 1]

Surroundings and location of vendors

Hawkers contributed to around 90% of contamination among panipuri vendors as compared to shops (50%). Those hawkers or shops located in

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crowded locations were more susceptible to contamination (90%). 71% of the vendors have fly breeding places nearby; among them, 84% are contaminated. The majority of the panipuri vendors (76%) are exposed to open air with no protective sheets or covering, which increases the chance of rainwater contamination. 85% of these vendors with no protective covering were contaminated with pathogenic organisms. There is a common practice of not keeping soap or hand wash for both vendors and customers. 90% of vendors with open dustbin were contaminated as compared to those with closed dustbin (50%). Waste collected in their personal dustbins and were later handed over to the municipality collection team, the next morning. Thus, the site of the shop, type of shop and method of keeping waste bins nearby are significantly associated with bacterial contamination. [Table 2]

Source and storage of elements of panipuri

Most (88%) of the vendors use can-water for preparation of panipuri, among them 86% are contaminated as compared to those using public tap water, though it is used by only 3% of vendors. Puri self-produced by the vendors, keeping the puri for more than 12 hours, and keeping the food containers open were more prone for contamination and this association was statistically significant. Vendors using re-usable plates (79%) had more chance of contamination. [Table 3]

Personal hygiene, behaviour and dressing pattern of vendors

Although only 44% of the vendors were having unclean dressing pattern, among them almost all (97%) were contaminated. More than 90% of vendors were not wearing gloves and aprons. The bacterial contamination is more prevalent among the panipuri vendors who maintain poor personal hygiene and have long nails. The rate of contamination was higher among those vendors who cleaned the work area and utensils occasionally and also using only a single bucket of water for the purpose of cleaning. The dressing of vendors, use of gloves, overall personal hygiene, condition of the vendor's nails, frequency of cleaning hands, and frequency of cleaning the working area were found to be highly significant. [Table 4]

Table 1: Association of demographic variables with the rate of bacterial contamination in Panipuri (N = 80)

Attributes (vendors)	Category	Contaminated n = 67 (%)	Non-contaminated n = 13 (%)	Total	P Value
Age (in years)	15-30	18 (69)	8 (31)	26	0.015*
	≥30	49 (91)	5 (9)	54	
Sex	Male	63 (84)	12 (16)	75	0.814
	Female	4 (80)	1 (10)	5	
Education	Illiterate	32 (94)	2(6)	34	0.031*
	Literate	35 (76)	11(24)	46	
Years of experience (in years)	< 10	56 (92)	5 (8)	61	<0.001*
	≥ 10	11 (58)	8 (42)	19	
Working time (in hours)	4-8 hours	19 (83)	4 (17)	23	0.860
	> 8 hours	48 (84)	9 (16)	57	
Average number of customers per day	< 100	46 (84)	9 (16)	55	0.967
	≥ 100	21 (84)	4 (16)	25	
Peak time of sale	12-4 PM	18 (90)	2 (10)	20	0.312
	4-8 PM	49 (82)	11(18)	60	

*** P value significance < 0.05**

Table 2: Association of surroundings and location of shop on rate of bacterial contamination (N = 80)

Attributes (vendors)	Category	Contaminated n = 67 (%)	Non-contaminated n = 13 (%)	Total	P Value
Shop type:	Hawkers	62(89)	8(16)	70	0.002*
	Shop	5 (50)	5 (50)	17	
Site of shop:	Crowded	46(90)	5(10)	51	0.038*
	Not crowded	21(72)	8(28)	29	
Surroundings of shop/ vehicle:	Poor	21(81)	5(19)	26	0.399
	Average	23(92)	2(8)	25	
	Good	23(79)	6(21)	29	
Any Drainage nearby:	Yes	33(85)	6(15)	39	0.838
	No	34(83)	7(17)	41	
How the waste bin kept?	Closed	6(50)	6(50)	12	0.001*
	Opened	61 (90)	7 (10)	68	

*** P value significance < 0.05**

Table 3: Association of source and storage of elements of panipuri on rate of bacterial contamination (N=80)

Attributes (vendors)	Category	Contaminated n = 67 (%)	Non-contaminated n = 13 (%)	Total	P Value
Water used for production	Can-water	60(86)	10(14)	70	0.223
	Bore well water	5(71)	2(29)	7	
	Public tap water	2(67)	1(33)	3	

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Mode of puri production	Self	54 (89)	7(11)	61	0.038*
	Agency	13(68)	6(32)	19	
Period of utilization of puri	<12 hours	5 (56)	4(44)	9	0.015*
	> 12 hours	62(87)	9(13)	71	
Did he kept food containers closed?	Yes	19(70)	8 (30)	27	0.021*
	No	48(91)	5(9)	53	

*** P value significance < 0.05**

Table 4: Association of food hygiene practices with the rate of bacterial contamination in Panipuri (N = 80)

Attributes (vendors)	Category	Contaminated n = 67 (%)	Non-contaminated n = 13 (%)	Total	P Value
Dressing of vendor	Unclean	34(97)	1(3)	35	0.004*
	Clean	33(73)	12(27)	45	
Use of gloves	Yes	2 (40)	3 (60)	5	0.006*
	No	65 (87)	10(13)	75	
Use of Apron	Yes	2(67)	1(33)	3	0.414
	No	65(84)	12(16)	77	
Personal hygiene	Poor	30 (97)	1(3)	31	0.006*
	Average	22(85)	4(15)	26	
	Good	15(65)	8(35)	23	
Condition of vender's nails	Long	28 (93)	2(7)	30	0.036*
	Short	25(86)	4(14)	29	
	Trimmed	14 (67)	7(33)	21	
Frequency of cleaning hands	Very frequently	16 (76)	5(24)	21	0.028*
	Frequently	21 (75)	7(25)	28	
	Occasionally	30 (97)	1 (3)	31	
Frequency of cleaning the working area	Very frequently	7(78)	2(22)	9	0.003*
	Frequently	18 (67)	9(33)	27	
	Occasionally	42 (96)	2 (4)	44	
Frequency of cleaning utensils	Very frequently	9(75)	3(25)	12	0.314
	Frequently	34(81)	8(19)	42	
	Occasionally	24(92)	2(8)	26	
Method of cleaning the utensils	Single bucket	49 (92)	4(8)	53	0.019*
	Changing water	16 (67)	8(33)	24	
	Running water	2(67)	1(33)	3	

*** P value significance < 0.05**

DISCUSSION

A total of 160 samples, including both masala pani and puri were analysed for the presence of pathogenic organisms during this study. This study showed that the overall bacterial contamination was 84%, with a high proportion of Micrococci, Bacillus spp., Klebsiella spp., Escherichia coli, and Citrobacter spp. These results are closer to the study done by Mehta H. D. et. al. in Morbi city, Gujarat in 2019 with 40 samples of panipuri, which showed 82% contamination with majority organisms being Escherichia coli, Staphylococcus aureus, Klebsiella spp., and Pseudomonas spp.[9] In a similar study done by Satish V. et al. in Bengaluru during 2021 with 100 panipuri samples, 96% of them had bacterial contamination, with the major organisms being Escherichia coli, Klebsiella spp., Enterococcus spp., and Candida.[11] Raghupathi C et al. evaluated the quality of pani puri in bidar during 2021 and reported that 73.75 % of the samples (a total of 80 samples) were contaminated by different pathogenic bacteria, which contain Escherichia coli, Staphylococcus aureus, Klebsiella spp., Pseudomonas spp., Bacillus spp., Salmonella spp., and Streptococcus as common isolates.[12] Yadav N. P. et al., Tambekar D. H. et al., V Teegala et al., and Khadka S et al. found bacterial contamination in pani-puri samples to be 70%, 93%, 74% and 54.6% respectively. Escherichia coli, Staphylococcus aureus, Klebsiella spp., and Pseudomonas were major bacterial contaminants in these studies.[2,4,13,14] Bacterial contamination is more prevalent in south and north zones of the Kalaburagi city, which were more crowded. In this study, those vendors with an age greater than or equal to 30 years are prone to contamination (91%), this may be due to poor adherence to hygienic practices. Although most of the vendors are literate (58%); a lack of proper knowledge on food handling, sanitation, and waste

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disposal were found to be major contributors to bacterial contamination of 94% among illiterates (42%). The lack of experience in making and serving panipuri also led to contamination (92%). Khadka S et al. reported 59.55% of vendors were illiterate; lack of awareness about food laws and regulation and lack of formal training were found to be important factors in food contamination as found by Mehta H. D. et al.[9,14]

In our study, hawkers with temporary makeshift stalls are more susceptible to around 90% contamination as compared to shops (50%). About 64% of vendors were located in crowded areas were more exposed to dust and flies, Mehta H. D. et al., Raghupathi C et al., Yadav N. P. et al and Tambekar D. H. et al. also found 74%, 44%, 42% and 73% of vendors in crowded areas.[2,4,9,12]

This study showed that keeping the waste bins open to flies can lead to an increase in breeding places and a higher risk of contamination (90%). Satish V et al. demonstrated that hawkers' direct exposure to the surrounding environment, insufficient dust protection, and their migratory patterns could be the main causes of significant contamination. The contamination is more likely to occur among those who use personal dustbins, which are kept open nearer to the preparation site. [11] In our study around 90% uses can-water for the production of masala pani and puri and it is responsible for 86% of bacterial contamination. They use cups without handles for fetching water, they also store the water in containers that are not cleaned properly and used to refill same container without washing. The contamination is more (89%) among the vendors, who produce puri by themselves and most probably due to mixing the flour with bare hands, keeping the prepared puri on the uncleaned surface, transporting it openly in plastic containers, and keeping the puri in an open box or plastic covering in the shop or hawkers. The majority of vendors keep puri for more than 12 hours, which provides more exposure to microorganisms (87%) and compared to less than 12 hours (56%). The vendors keeping containers with masala pani, chopped onion, and other masala items open, where found to have 91% of contamination. Poor dressing and personal hygiene by vendors increased the chance of contamination (97%). Those vendors with non-trimmed nails were predisposing to high chance of contamination (90%). Khadka S et al. revealed 79.5% were not using gloves, 70.3% were using open food containers and 69.7% did not trim the nails regularly.[14]

In our study the vendors were using the same towel after washing their hands, which could be the probable source of contamination. Those vendors were used to clean the work area, chopping plate, knife, and surrounding area occasionally were found to have more (96%) contamination than those cleaning frequently (67%). Metha H. D. et al. showed that 80% of vendors were having inadequate personal cleanliness. He also identified that poor hand hygiene, handling the food with bare hands and unhygienic serving practice were the major source of contamination.[9] Tambekar D. H. et al. highlighted that bacterial contamination is more prevalent among vendors who less frequently washing hands, clean the cooking surface, clean the utensils, and use a single bucket for cleaning utensils. The vendors used to wash hands once or three times a day and use the same hands, which are used for peeling potatoes, cutting vegetables, and buying cash from customers. Only a few vendors wear disposable gloves, an apron, and head gear.[2]

CONCLUSION

The panipuri vendor's personal hygiene, poor handling of food without gloves, washing utensils less frequently without soap, and multiple contacts with food and other surfaces, including cash and cooking surfaces, are responsible for the contamination of the food. The isolation of the afore-mentioned organisms is not acceptable under the minimal standards of safe and wholesome drinking water and food safety standards, which might lead to the spread of infection among the population.

Recommendations

The most effective step in preventing food contamination is to ensure adherence to strict hygienic practices by both consumers and vendors. If we want to provide safe and hygienic street food, it can be achieved by using stainless steel utensils and work areas, masala pani and puri vending machines, and disposable plates and cups (for masala pani), which prevent the frequency of contact with bare hands. The widespread contamination highlights the need for health education initiatives on food safety for both food handlers and consumers, as well as for standardized food inspection. This also necessitates the need of safe and wholesome drinking water supply and a proper waste disposal system. Achieving a head start in this fight against food contamination will require further research and the development of an efficient vaccinations against food-borne infections.

Limitation

This study was not able to focus on laboratory investigations of food handlers, like serological test, skin and nail conditions and stool samples, who themselves might be the source of contamination.

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Conflict of Interest

The authors declare no conflict of interest.

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