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BACTERIAL ANALYSIS AND SURVEY OF THE STREET FOOD OF KATHMANDU IN RELATION TO CHILD HEALTH

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ABSTRACT

Analysis of street foods of Kathmandu for bacterial contamination was performed in 12 different street foods. The surveillance study was carried in 200 children of primary grade from public school and 12 street vendors for the health hygiene and hazards associated with street food. Poor hygiene practice in preparation and handling of street food has been observed in the vendors. The lack of the knowledge in vendors about the source of bacterial contamination and absence of surveillance on street food has subjected street food to the high potential for food borne illness. The inadequate safety measure adopted by the targeted consumers of street food, the children, has augmented the risk associated with street food. All the food samples analyzed were contaminated with bacteria. The mesophilic count was recorded highest in *Panipuri* while as coliform count was highest in *Chana tarkari*. The least count of both was observed in *Aaloo chop* . Highest number of *Staphylococccus aureus* was found in *Kerau* ($1.5X10^3$cfu/g) and lowest in *Momo* (8.3 cfu/g). The dominant bacteria contaminating the food was *S. aureus* followed by *Bacillus alvei, Escherichia coli, Enterobacter aerogenes, Bacillus subtilis, Serratia* sp., *S. saprophyticus*. The contaminated hand and clothing of the person who prepare food are the major source of *S. aureus*. Highest percentage of *E. coli* found in *Panipuri* must be due to the use of contaminated water. *Chana chatpate* and *Chana tarkari* were the foods found to be contaminated with *Salmonella* sp. The type of food and the degree of hygiene practice adopted by vendor reflects the type and magnitude of bacterial contamination. Implementation of hygienic practices in vendors may reduce the contamination of street food and health education of the school children will curtail the incidences of food borne illness. Periodical monitoring of quality of street food will avoid any future outbreaks of bacterial pathogen.

Key words: vendors, coliform count, *S. aureus*, *E. coli*, food borne illness

INTRODUCTION

Street foods, as defined by Food and Agriculture Organization, are a 'ready to eat food and beverages prepared and/or sold by vendors and hawkers in the street and public places (FAO 1997). It is consumed each day by an estimated 2.5 billion people worldwide (FAO 2007). Though preparation and sale of food on street is an old practice in developing countries, the urbanization has augmented the habit of consuming street food. These cheaper foods are popular among the person who have limited time and wants to save money for surviving in the city. Millions of people consume snacks, meals and drinks sold by street vendors in the developing countries (FAO 1998). The street foods have not only been limited within low income people but people from various walks of life have been found to relish street food.

Despite being popular, cheap and readily available the street foods are associated with serious health problems. Unhygienic preparation and handling, selling the foods road side, insufficient

supply of water for cleaning purpose make street food one of the major sources of food borne illness (Das *et al*. 2010). Food borne illness mainly affects the gastrointestinal tract and the potential bacterial pathogens associated are *E. coli*, species of *Salmonella*, *Shigella* and *S. aureus* (Buchmann *et al*.1999, Barro *et al*. 2006, Tambekar *et al*. 2009).

The growing population of dwellers in Kathmandu has increased the demand for street food and as such there has been increase in the number and varieties of food sold by the vendors. However, less attention has been paid to the health and safety of the consumer. In most places the street foods are sold open in unhygienic surroundings with houseflies, fruit flies and air-borne dust as the source of contamination. Openly sold fruit juices have shown to be potential sources of bacterial pathogens such as *E. coli* 0157:H7, species of *Salmonella, Shigella* and *S. aureus* (Buchmann *et al*. 1999, Sandeep *et al*. 2001). The consumers on the other hand have not been aware about the serious problem associated with street food. Children are the most vulnerable group of people who are at the risk of health problem associated with food. These prime consumers of street food are easily lured by the cheap street food. As such the street food vendors are generally found near the areas of the school in Nepal. Street food have become so popular in Tanzania that two third of the school children buy food from street vendor every day (FAO 2007).

Reckless handling and preparation of street food leading to contamination of food is a serious problem in our country. An effective surveillance and careful monitoring of food at different stages such as producing, transporting, handling, storing, processing and marketing need to be implemented. Considering the health of the children an attempt is made to isolate and identify the microorganisms associated with street foods and survey the practices of selling street food and its consumption.

MATERIALS AND METHODS

Collection of Sample

The food samples were collected from street vendors of Kathmandu metropolitan city in the area around the primary school adopting the random sampling method. Clean spoon dipped in 95% ethanol and flamed was used to collect each food sample and carried to the laboratory in a sterile plastic bag kept in ice chilled icebox.

Sample Processing

Homogenate of each food sample was prepared by aseptically transferring 25g of food into sterile blender with 225ml Buffered Peptone Water (BPW) and blending for 2 min (Padhye and Doyle 1991).

Enumeration and Identification of Bacteria

Aerobic mesophilic and coliform: Enumeration was carried out following the procedure described by Refai (1979). Serial dilution of the food homogenate was carried out up to 10^{-6} dilution in BPW. Pour plate technique was employed for each dilution in sterile Plate Count agar (PCA) to enumerate total mesophilic bacteria. Similarly, sterile Violet Red Bile agar (VRBA) was used to enumerate coliform bacteria. Both agar plates were incubated at 37° C for 48h.

Staphlylococcus aureus: Spread plate technique was employed for the detection. 0.1ml of each sample dilution was spread on Baird-Parker Egg Yolk Tellurite agar plate and Mannitol Salt agar plates individually with sterile glass rod and incubated at 37° C for 24h. Black and shiny colonies with narrow white margins from Baird-Parker Egg Yolk Tellurite agar plates and bright yellow colonies from Mannitol Salt agar plates were picked as presumptive positive for *S. aureus*. The colonies were further confirmed by biochemical tests.

Salmonella sp.: The food homogenate was transferred into tetrathionate broth and incubated at 37° C for 24h for pre-enrichment. The culture from the broth was streaked on sterile Brilliant Green agar plates and incubated at 37° C for 24h. Pinkish white colonies surrounded by bright red medium were picked and further confirmed by biochemical tests.

The suspected organisms from corresponding selective or differential medium for coliforms, *S. aureus* and *Salmonella* sp. were sub-cultured in Nutrient agar plates. The isolated bacteria were identified on the basis of colonial characteristics, morphological and biochemical properties following the “Standard microbiological methods” described by Bergey’s Manual of Systematic Bacteriology (1985).

Biochemical tests: Biochemical tests were performed respective to the suspected organism. Catalase, Oxidase, Coagulase, Urease, Gelatin liquefaction tests were carried out for *S. aureus*. Similarly, Catalse, Oxidase, IMViC, TSI, Urease tests were performed for *E. coli* and *Salmonella* sp.

Sterility of the medium was checked by incubating uninoculated broths and plates when used simultaneously for the quality control of the tests.

Surveillance Study

Primary school children and street vendors were interviewed randomly for the information on the hygienic practice in preparation and handling of street food and history of disease possibly caused due to street food consumption.

RESULTS AND DISCUSSION

Street food appears to be on the rise encouraged by growing urban populations and public demand for readily available cheap food. However, the generally unregulated street food tends to observe poor hygienic practices and breed significant public health problems (Arambulo *et al*. 1994). At various times street foods have been found to harbor organisms responsible for causing typhoid, brucellosis, food poisoning, dysentery and diarrhea. The street vendors are mostly uninformed of good hygiene practices, which can increase the risk of street food contamination (Bhaskar *et al*. 2004). Therefore, there is high potential health risk associated with initial contamination of foods by pathogenic bacteria as well as subsequent contamination by vendors during preparation, handling, and cross contamination (Mosupye and van Holy 2000).

The survey was conducted in 12 street vendors near the school area to study the sanitary condition. Most of them do not practice safe method of storage of food. The limited storage space restricted them from separately storing cooked and uncooked food.

In most cases, running water is not available at the site of selling so the water used to cook

food and wash is mostly stored in traditional pot '*Gagro*', buckets or tin container. This could limit the proper washing of the hands and utensils. Though most of the vendors confessed that they clean the utensils, it was not observed but based on the interview. Although detergent was used for cleaning purpose, use of ash was still found to be practiced and very few mix ash and mud.

Hand washing practice though most of the vendors claimed of adopting were only restricted using plain water. Very few were found to use soap and water for hand washing.

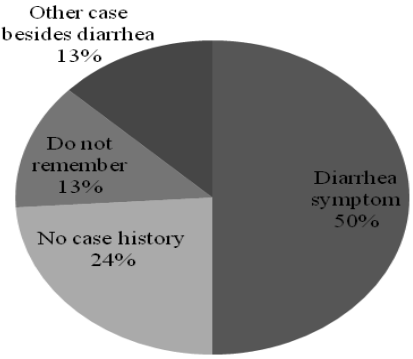

Figure 1. Diarrhea associated health hazards in school children consuming street foods.

Most of the vendors cook food beforehand and are generally not efficiently protected from flies. In some cases the food were covered by lid or net to prevent flies. However safe food storage temperature is not applied. About 96% of the vendors sold their food by the same day, while 4% said the food remain. Most of the leftover were kept for the next day while some times they were eaten up or thrown away depending upon the situation. Stale and improperly stored foods are highly potential for food-borne illness. Children are generally unaware of the risks associated with consuming the street food so they are highly vulnerable to outbreaks of food borne illness.

The results from the surveillance of 200 primary school children from grade 1 to 5 of the age between 5-14 years showed that the children of higher grade buy street food compared to that from the grade 1 who brings lunch from home. Besides, in the lower grade children, higher percentage of hand washing practice has been found compared to the higher grade children. As the children grow up they were found more negligent towards their health. The survey revealed that 50% of the students had encountered with diarrheal symptoms while 24 % did not have any history of disease related to consumption of street food (Fig. 1).

A total of 12 different food samples were analyzed (Table 1). All the samples were found contaminated with bacteria. The highest number of mesophilic count was observed in *Panipuri* and lowest in *Aaloo chop* (Table 1). Similarly the total coliform count was highest in *Chana tarkari* and lowest in *Aaloo chop*. Since different food samples were taken from different vendors the

lowest incidence of both mesophilic and coliform count must be attributed to the difference in the hygienic condition of the vendor.

Table 1. Total plate count and coliform count of food samples.

Sample	Type of food	Total plate count (cfu/gm)	Coliform count (cfu/gm)
1.	Potato curry	$8.83X10^5$	$5.8X10^2$
2.	*Chana chatpate*	$48X10^5$	$9X10^1$
3.	*Momo*	$80X10^5$	$5X10^1$
4.	*Chana* fried	$100X10^5$	$6X10^4$
5.	*Khuwa baraf* (ice-cream)	$260X10^5$	$1.76X10^2$
6.	Orange juice	$67X10^5$	$3X10^5$
7.	*Kerau tarkari*	$12X10^5$	$7.6X10^2$
8.	Egg curry	$35X10^5$	$4X10^5$
9.	*Chana tarkari*	$51X10^5$	$1.8X10^6$
10.	*Panipuri*	$101X10^5$	$5X10^5$
11.	Soda	$13X10^5$	$6.03X10^2$
12.	*Aaloo chop*	$2.6X10^5$	$1.7X10^1$

Cfu: Colony forming unit.

The highest number of *S. aureus* was found in *Kerau* ($1.5X10^3$cfu/g) and lowest in *Momo* (8.3 cfu/g). No growth was found in *Chana-fried, Khuwa baraf, Panipuri* and Soda (Table 2).

The presence of *E. coli* was shown in *Panipuri,* potato curry, *Khuwa baraf, Kerau tarkari,* egg curry and momo (Table 3). The main source of contamination of *E. coli* might be through contaminated water supplies (Tambekar *et al*. 2009). The outbreak of *E. coli* can result from using contaminated water in food as well as for washing utensils. Infection with enterotoxigenic *E. coli* (ETEC) is a frequent cause of diarrhea in developing countries (Weekly report 1994). *E. coli* outbreak occurred in four elementary schools in July 1996 affecting more than 800 people in Japan, eating tuna paste served for lunch (Mitsuda *et al*. 1998).

Table 2. Total *S. aureus* found in street foods.

Food sample	Mean *S. aureus* count (cfu/gm)
Potato curry	$1.36X10^3$

Chana chatpate	$4X10^{1}$
Momo	8.3
Chana fried	0
Khuwa baraf	0
Orange juice	$1.3X10^{2}$
Kerau tarkari	$1.5X10^{3}$
Egg curry	$1.3X10^{3}$
Chana tarkari	10
Panipuri	0
Soda	0
Aaloo chop	10

Cfu: Colony forming unit.

The two food samples, *Chana chatpate* and *Chana tarkari* were found to be contaminated with *Salmonella* sp. (Table 3). Several incidences of *Salmonella* associated outbreaks of food poisoning have occurred around the world. *Salmonella enteritidis* was isolated from the food with egg as an ingredient in New York in 1985 (Morse *et al*. 1994), from ice-cream, cream cakes, mayonnaise in Poland between 1961-1991 (Glosnicka and Kunikowska 1994). *S. newport* was isolated from outbreak of food poisoning that occurred in 1991 among students of the Gonder College of Medical Sciences, Ethiopia (Aseffa *et al*. 1994).

Table 3. Incidences of *Salmonella* sp. and *E. coli* in food samples.

Type of food	**Incidence of *Salmonella* sp.**	**Incidence of *E.coli***
Potato curry	0(0%)	1(33%)
Chana chatpate	1(33%)	1(33%)
Momo	0(0%)	1(33%)
Chana fried	0(0%)	0(0%)
Khuwa baraf	0(0%)	1(33%)
Orange juice	0(0%)	0(0%)
Kerau tarkari	0(0%)	1(33%)
Egg curry	0(0%)	1(33%)
Chana tarkari	1(33%)	0(0%)
Panipuri	0(0%)	1(33%)

Soda	0(0%)	0(0%)
Aaloo chop	0(0%)	0(0%)

Among the organisms isolated from food samples, *S. aureus* (11.76%) was the most dominating followed by *Bacillus alvei* (9.80%), *E. coli* (7.84%), *Enterobacter aerogenes* (7.84%), *Bacillus subtilis, Serratia* sp.*, S. saprophyticus,* etc. (Table 4). The maximum contamination with *S. aureus* reflects the poor hygienic practices. The contaminated hands and dirty clothing must be the major source of contamination during handling and preparation. Predominant strains of *S. aureus* were recorded in fish samples by Sokari (1991).

However, all the bacterial organisms isolated from the food samples were not pathogens. Some of them are opportunistic and non pathogens. These organisms present in the dust, air and soil around the place where the foods are being sold can be carried by wind or flies.

The hygienic setting for selling of street food will always be of poor standard. Besides, prohibiting the street vendors from selling food is almost impossible. Therefore, the government health agencies must adopt measures to educate the vendors on food safety and hygienic practices and enforce proper guidelines for street food vending. On the other hand the school children must be educated about the relation of street food and the food borne illness as well as good hygienic maintenance. Apart from that, regular monitoring of the quality of street food for consumption must be implemented for the safety of the general public.

Table 4. Frequency and percentage of different bacterial organisms detected in food samples.

Bacterial species	**Frequencyof detection**	**Percentage**
Bacillus alvei	5	9.804
Bacillus cereus	1	1.96
Bacillus licheniformis	2	3.921
Bacillus megaterium	1	1.96
Bacillus subtilis	3	5.882
Citrobacter diversus	1	1.96
Citrobacter fruendii	1	1.96
Enterobacter aerogenes	4	7.843
Escherichia coli	4	7.843
Flavobacterium sp.	1	1.96
Klebsiella oxytoca	2	3.921
Klebsiella pneumonia	2	3.921
Micrococcus luteus	2	3.921

Micrococcus sedentarius	1	1.96
Pediococcus	1	1.96
Proteus mirablis	2	3.921
Pseudomonas aeruginosa	2	3.921
Pseudomonas stutzeri	1	1.96
Serratia sp.	3	5.882
Staphylococcus aureus	6	11.765
Staphylococcus haemolyticus	1	1.96
Staphylococcus hyicus	1	1.96
Staphylococcus saprophyticus	3	5.882
Salmonella sp.	1	1.96

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