Microbiology



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ABSTRACT

The increase in cases of food borne illnesses related to street vended fruit salad in developing countries is of serious public health concern. Ready-to-eat salads are considered as a high-risk food because they do not require any heating, was bing or cleaning prior to concurrent in This study was therefore conducted on street worded fruit salad to determine

washing or cleaning prior to consumption. This study was therefore conducted on street-vended fruit salad, to determine their microbial quality using microbial limit test. A total of 10 different salads were collected aseptically from different vendors of Mumbai, India. The homogenized material (10g) was serially diluted and inoculated on nutrient and MacConkey agar by Surface-Spread Plate technique. The total bacterial count falls within the range of 1.70 × 105 and 2.40 × 105 cfu/g, with sample 7 showing the highest count of 2.40 × 105 cfu/g and sample 1 showed the lowest count of 1.70× 105 cfu/g. Bacterial growth was observed in all the food types; the most prevalent bacteria were Aeromonas hydrophilla (28.8%), Citrobacter freundii (19.2%), Salmonella sp (15.3%), Pseudomonas aeuroginosa (13.4%) , E.coli (11.5%) and Bacillus (11.5%). This study revealed the potential hazard of ready-to-eat salads and it is the need of the hour to perform a surveillance study at national scale.

KEYWORDS : Ready to eat fruits, vendors, hygiene, coliform, contamination

Introduction

Ready-to-eat fruits are fruits that can be bought directly from street vendors or hawkers or at local markets and eaten immediately i.e. without necessarily having to cut, peel or rinse them before consumption as they have already been prepared by the vendors (Oranusi and Olorunfemi 2011). Fruits are an extraordinary dietary source of nutrients, micronutrients, vitamins and fibre for humans and are thus vital for health and well being.

Fruit salad is usually a combination of various fresh fruits such as apples, water melon, pineapples, cucumber, pawpaw and orange. They are usually sliced into small pieces and packaged in small transparent plastic bowls. The salad can be eaten using fork or tooth pick, with or without milk added to it (Kalia and Gupta 2006). According Edward *et al.* (2012), fruit salad is low in cholesterol, sodium and saturated fat but high in vitamins A, C and D, manganese, copper and dietary fibre.

The different fruits used in preparing the salad are usually kept on the ground near the slicing tables without any form of protection. Fruits are prone to microbial contamination because they are constantly in contact with soil, dust and water, and by handling at harvest or during post harvest processing. Pathogenic microorganisms may also enter the fruits through damaged surfaces, such as punctures, wounds, cuts and splits. Such pathogens may become internalized, survive and grow within the fruit and consequently become health hazard to consumers (Brooks 2014).

The increased consumption, coupled with the associated risk of disease to which consumers may be exposed, is a matter of great concern. Moreover, the case is worsened by the fact that sliced fruit street vending is done without adequate storage conditions, thereby exposing the sliced fruits to flies and other disease-causing agents (FDA 1999a).

Salmonella sp has been reported to survive and grow rapidly on water melon held at room temperature and the level of contamination did not change when the melon was stored at refrigeration temperature (Brooks 2014). Outbreaks of listeriosis and salmonellosis have also been associated with the consumption of readyto-eat fruit salad (Mensah et al., 2002). Bacteria like *Salmonella spp., Shigella spp., Campylobacter spp. and Escherichia coli* can contaminate sliced fruits through contact with sewage and contaminated water (Jones 1990). This study was designed to assess the microbial contaminants of ready to eat fruits sold in Mumbai, India, in order to highlight the health implications of consuming such unwholesome ready to eat fruits.

Materials and Methods

Samples were collected from ten different locations, local market in Mumbai. These ten vending sites were chosen because these markets

are the major one in town and many vendors patronize the market for sales. All the samples were collected wholly in small transparent plastic bowls as sold and transported in ice-packed cooler to the laboratory where they were analyzed within 1 hour after collection. The samples were analyzed according to the FDA (2001).

Isolation and Enumeration of Bacteria. Nutrient agar and Luria burtini broth were prepared and sterilized by autoclaving at 121°C for 15 mins. 10grams of each fruit salad sample was blended in a sterile blender with 90mls of peptone water for two minutes. 10 fold serial dilutions were prepared and 1ml of each dilutions was inoculated on media. The plates were then incubated at 37 °C for 24-48 hours and examined for colony formation. After incubation, colonies were counted using a colony counter, while pure cultures of isolates were obtained by sub-culturing in fresh medium using the streak plate method.

Identification of isolates. The bacteria isolates were identified based on standard methods of Speck (1976). Isolates were Gram stained and specific biochemical tests were performed to identify the isolates.

Results and Discussion

The present investigation reveals high microbial load in the fruit salad. The microbial contaminants consist of comprising different pathogens such as *Escherichia coli, Staphylococcus aureus, Bacillus cereus, Salmonella sp., and Pseudomonas sp.* Most of the bacteria isolated are almost always readily available in our environment. The presence of these organisms can be linked to a number of factors such as improper handling and processing, use of contaminated water during washing, cross contamination from other fruits and vegetables or the use of dirty processing utensils like knives and trays (Oranusi and Olorunfemi 2011). Bryan et al., (1992) and Beuchat, (1996) isolated species of bacteria which include *Bacillus spp., Salmonella spp and Escherichia coli* from street vended foods and the presence of these organisms were thought to be as a result of inappropriate processing.

Table 1: The microbial count of the fruit salad samples

Sample	Total viable count	Total coliform count	
	CFU/g 10⁵	CFU/g 10 ³	
Sample 1	1.7	3.33	
Sample 2	2.6	3.67	
Sample 3	2	4.67	
Sample 4	1.93	0.43	
Sample 5	1.83	1.67	

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Sample 6	1.97	4
Sample 7	2.4	4.33
Sample 8	2.3	3.37
Sample 9	1.73	1
Sample 10	2.1	2.13

From the results obtained, the total bacterial count falls within the range of 1.70×10^5 and 2.40×10^5 cfu/g, with sample 7 showing the highest count of 2.40×10^5 cfu/g and sample 1 showed the lowest count of 1.70×10^5 cfu/g (Table 1). It is assumed that most of these bacterial isolates have the capacity to cause disease.

The high coliform count is very disturbing with the least count of 0.43 $\times 10^3$ cfu/g (Table 1). It is important to observe that the presence of *E. coli* as an enteric bacteria with established pathogenicity in man poses a risk to young children, the elderly and the immunocompromised persons if as low as $10^1 - 10^2$ /g(infective dose) is found (ICSMF 1974).

Bacterial growth was observed in all the food types; the most prevalent bacteria were Aeromonas hydrophilla (28.8%), Citrobacter freundii (19.2%), Salmonella sp (15.3%), Pseudomonas aeuroginosa (13.4%), E.coli (11.5%) and Bacillus (11.5%) (Table 2). This is consistent with the findings of previous studies. The bacteria found to spoil the fruits were, Klebsiella, Bacillus, Escherichia coli, Staphylococcus, Pseudomonas, Salmonella and Lactobacillus (Nwachukwu and Chukwu 2013). The presence of these bacteria may be due to the unhygienic environmental conditions and poor handling. The results of this experiment suggest that decaying fruits nourishes various types of bacteria which may cause harm to the consumers.

Colonization of fruits and vegetables by the invading microorganism is a critical phase in the microbial spoilage of produce. The colonization process involves the ability of the microorganism to establish itself within the produce and degrade certain specific cell wall polymers such as protopectin, the cementing substance of the produce. The magnitude of the symptoms of the induced disease is a reflection of the extent of colonization (Chuku *et al.*, 2008).

The occurrence of enteropathogenic bacteria such as *E. coli, V. cholerae* and *S. typhi* on the foods examined renders them as vehicles for food – borne infections. *E. coli* if consumed in foods is liable to causing peritonitis, mastitis, septicemia and gastrointestinal infections sometime becoming fatal.

World Health Organization. (2002) reported that *Salmonella* spp. causes Salmonellosis and typhoid fever and *E. coli* O157:H7 causes severe illness and deaths, especially among children in several countries. The chances of contamination is heightened by the fact that sliced/peeled fruit street vending is done without adequate storage conditions, thereby exposing the sliced fruits to flies and maintains the produce at optimum temperatures for invasion and proliferation of contaminants, pathogenic mesophiles and other disease-causing agents. Poorly processed street vended produce have been identified as an important cause of diseases in developing countries. Fruits have been associated with outbreaks of food-borne disease in many countries; organisms involved include bacteria, fungi, viruses and parasites.

Isolates	Total number	%
Citrobacter freundii	10	19.2
E.coli	6	11.5
Pseudomonas aeuroginosa	7	13.4
Salmonella sp	8	15.3
Bacillus	6	11.5
Aeromonas hydrophilla	15	28.8

Conclusion

The present study revealed bacterial contamination in street-vended fruit chats. Handling practices carried out by street vendors and the prevailing conditions in which these products are displayed is the main cause of contamination. The levels of contamination clearly suggested the potential risk of food borne out-breaks. Since, no international or local standards exist in the country, standardization of health and safety status of such products must be arbitrated by appropriate regulatory authorities which can minimize the risk of bacterial diseases to a great extent.

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