Efficacy of Commonly Used Decontaminants on Bacterial Contamination on Food Garnishes

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Abstract- Food garnishes are often used in an uncooked form. Hence, the chance of contamination is high. The present research work was carried out to isolate potentially pathogenic and food spoilage causing bacteria from commonly used food garnishes. Six such bacteria were isolated and characterized and antibiotic sensitivity tests were conducted. It was found that it is concluded that the most dominant bacteria on food garnishes was *Pseudomonas aeruginosa* followed by *Escherichia coli, Staphylococcus aureus, Salmonella sp., Klebsiella sp., and Citrobacter sp.*. The combination of vinegar and salt was found to be more effective against contaminant bacteria isolated from food garnishes.

Keywords: Food garnishes, Bacterial contamination, Decontaminants

INTRODUCTION

A food garnish is an item or substance used as a decoration or ornamentation accompanying a prepared food dish. In many cases, it may give added or contrasting flavor. Some garnishes are selected mainly to augment the visual impact of the plate, while others are selected specifically for the flavor they may impart. A food item which is served with garnish may be described as being garni, the French term for 'garnished'). Garnishes can be broadly classified into edible and non-edible garnishes. Edible garnishes are those which can be consumed along with the food to which it is added, for example, coriander on Indian curry dishes. Non-edible garnishes aid in the visual impact of the food and they do not impart any flavour to the food. These are usually discarded by the consumers, example: cocktails can be garnished with plastic cocktail umbrellas.

Edible garnishes make food items look more appealing, complement the food, add nutritional value, colour balance and enhance the taste of the food. Some of the edible garnishes include cloves, cardamom, curry leaves, chilli pepper, kasuri methi, mint, pepper corns, oregano powder, coriander leaves, spring onions, paprika powder, tomato, pomegranate, coconut, dry fruits, dry chilli, grapes, honey, lime etc. Along with these garnishes many other food garnishes are used worldwide in various food delicacies (Goldstein, 1999).

Food contamination refers to the presence of harmful chemicals and microorganisms in food, which can cause consumer illness (Heiser and Simmonds, 1976). Food-containing harmful microorganisms are responsible for more than 200 diseases ranging from diarrhea to cancers. Some other diseases caused by microbial contaminants are cholera, campylobacteriosis, *E. coli* gastroenteritis, salmonellosis, shigellosis, typhoid and paratyphoid fever, amoebiasis and poliomyelitis. Though numerous pathogens cause similar symptoms (such as diarrhea, abdominal cramps and nausea), the illness caused by foodborne pathogens vary with clinical manifestations, severity and duration. Most foodborne illnesses are more severe or prolonged and are limited to brief episodes of diarrhea, nausea or other acute gastrointestinal systems. Several other illnesses can result from the consumption of foods contaminated by microbial pathogens and include fever, vomiting, weakness, chills and aches, headaches, abdominal pain, constipation, sore mouth, blurred vision, muscle paralysis. A small portion of foodborne illnesses are severe or fatal, however complications such as septicemia, localized infection of other organs and spontaneous abortion in pregnant women are the most severe acute illnesses associated with foodborne pathogens that contaminate food.

Decontamination is the process of cleansing an object or substance to remove contaminants such as micro-organisms or hazardous materials. The purpose of decontamination is to prevent the spread of micro-organisms and other noxious contaminants that may threaten the health of human beings or animals or damage the environment. Methods of decontamination includes Physical cleaning, Water purification, Ultrasonic cleaning, Disinfection, Antisepsis and Sterilization whereas different agents of decontamination includes Commercial fruits and vegetable wash, Baking soda, Vinegar and Salt water.

Commercial fruits and vegetable wash: A vegetable wash is a cleaning product designed to aid in the removal process of dirt, wax and pesticides from fruit and vegetables before they are consumed. All fresh produce, even organic, can harbor residual pesticides, dirt or harmful microorganisms on the surface. Vegetable washes may either be of specially-marketed commercial brands, or they may be home recipes. Commercial vegetable washes generally contain surfactants, along with chelating agents, antioxidants, and other agents. Home recipes are generally dilutions of hydrogen peroxide or vinegar, the former of which may be dangerous at high concentrations (Bradley et al., 2011).

Baking soda: Baking Soda acts a decontaminant as well as cleaning agent because it is a mild alkali and can cause dirt and grease to dissolve easily in water for effective removal. When it is not fully dissolved, like when it is sprinkled on a damp sponge, it is mildly abrasive.

Vinegar: Vinegar is safe to use as a home remedy to clean, sanitize or surface sterilize a variety of fresh fruits and vegetables. However, the extent and effectiveness of sanitation by using vinegar will depend on the nature of the suspected disease-causing agents. In other words, fungi and bacteria can be effectively removed from these fresh products by using vinegar, but the effectiveness of the vinegar depends on which bacterium and/or fungus is on fruit or vegetable, the concentration of the vinegar, the temperature of the water and the amount of time it is exposed to the vinegar.

Salt: Utilization of salt allows for a comparable or higher reduction of the bacterial load from vegetables and fruits. It removes all the wax and dirt on it. Washing with salt water is very effective to kill the microbes.

Thus, besides the numerous beneficial aspects of food garnishes, there are also a few drawbacks. The drawback of food garnishes which is of concern to a microbiologist is the fact that food garnishes are added to a dish uncooked and thus, potentially capable of being vectors of microorganisms and cause food spoilage and food poisoning (Campbell et al., 2001; Elviss et al., 2009). A lot of work has been done on uncooked vegetables, spices and nuts (Ali et al., 2014; Andleeb et al., 2013). However, not much work has been done on garnishes so far, except parsley and mint (Hsu et al., 2010; Naimi et al., 2003; Wu et al., 2000), even though they have been a part of our daily food. Methods to improve the microbial quality of these food garnishes have also been minimally analyzed. Hence, having a better knowledge on food garnishes, the possible microorganisms they harbor, their types, and in turn the spoilage and poisoning they cause is essential. Thus, the scope of research on garnish-borne food spoilage and food poisoning causing microorganism and on measures to control them is enormous. Bacteria related food poisoning is the most common. More than 90 percent of the cases of food poisoning each year are caused by *Staphylococcus aureus, Salmonella, Clostridium perfringens, Campylobacter, Listeria monocytogenes, Vibrio parahaemolyticus, Bacillus cereus,* and Entero-pathogenic *Escherichia* etc (Zhuang et al., 1995).

In case food spoilage or food poisoning causing microorganisms are found to be resistant to a wide range of antibiotics, attempt should be made to reduce the chance of such contamination in the first place, especially for food garnishes which remain uncooked. This can be done by use of decontaminants to get rid of or to reduce the number of microorganisms present on garnishes before application on to food items. Apart from the general decontamination methods, for example heat, liquid disinfectants, and radiation, there are few home remedies, which can be practiced (Tzortzakis, 2010; Bradley et al., 2011). These home remedies include various organic acid, plant extracts etc. The advantage of using these home remedies as decontaminants is that they are safer, cheaper and eco-friendly. The objective of this study was to efficacy of commonly used decontaminants against antibiotic resistant bacteria on food garnishes.

MATERIALS AND METHODS

Sample Collection: Approximately 50-100g of unpacked garnish samples such as Coriander, Coconut, Spring onion, Curry leaves, Chilly pepper, Black pepper and Kasuri methi were collected in sterile aluminum foil pouches. Samples were transported to the laboratory and analyzed as soon as possible (Table 1).

Table 1: Garnish samples used in the study

Sr. No.	Food Garnishes	Scientific names
1	Coconut	*Coco nucifera*
2	Coriander	*Coriandrum sativum*
3	Spring Onion	*Allium fistulosum*
4	Curry Leaves	*Murraya koenigii*
5	Chilly Papper	*Capsicum annuum*
6	Black Pepper	*Piper nigrum*
7	Kasuri Methi	*Trigonella foenumgraecum*

Isolation and Identification of bacteria from Food garnishes: Different media like Mannitol salt agar, MacConkey agar and Pseudomonas Isolation agar were used for isolation of bacteria associated with food garnishes. On these plates samples of food garnishes were placed by using sterile forceps and needle were used for taking small pieces of food garnishes and plating them on to the media plates. The plates were incubated overnight at 37°C. Microscopic examination, cultural characteristics and biochemical characterization of the isolates were carried on the basis of characters given in the Bergey's manual (2012); (Collee and Marr, 1996).

Antibiotic Sensitivity test: Antibiotic sensitivity test was performed by Kirby Bauer Disc Diffusion method (Bauer et al., 1966). Six different types of antibiotics were used in the study (Table 2). Isolated bacteria were grown on nutrient agar at 37^0C for 24 hours and the colonies were suspended in sterile saline water equivalent to a 0.5McFarland standard (1.5X108CFU/ml). Hi-sensitivity agar plate was uniformly seeded by adding 100µl inoculated broth and was spread by means of spreader. The antibiotic discs were placed on each inoculated Hi-sensitivity agar plate. The plates were incubated at 37^0C for 18 hours. The diameter of the zone of inhibition was observed in mm and the isolates were classified as "resistant" or "sensitive" based on the standard interpretative chart according to Clinical and Laboratory Standards Institute (CLSI) guidelines (CLSI, 2007).

Table 2: Antibiotic used in the study

Antibiotics	Abbreviation	Concentration
Erythromycin	E	15mcg
Tetracycline	TE	10mcg
Levofloxacin	LE	5mcg
Gatifloxacin	GAT	30mcg
Azithromycin	AZM	15mcg
Chloramphenicol	C	30mcg

Decontamination test by well diffusion method: The following three types of decontaminants were used (Table 3). Six different combinations of decontaminants were prepared (Table 4). Isolated bacteria were grown on nutrient agar at 37^0C for 24 hours and the colonies were suspended in sterile saline water equivalent to a 0.5McFarland standard (1.5X108CFU/ml). Hi-sensitivity agar plate was uniformly seeded by adding 100µl inoculated broth and was spread by means of spreader. The wells of 6 mm diameter were cut into the agar medium with a sterilized cork borer. Then 20µl each of the decontaminant solution were added separately into the separate wells. The plates were incubated at 37^0C for 18 hours. The diameter of the zone of inhibition around each well was measured and recorded (Bauer et al., 1966).

Table 3: Decontaminants used in the study

Decontaminants	Abbreviation
Baking soda	BS
Salt	S
Vinegar	Vi

Table 4: Six different combinations of decontaminants used in study

Sr. No.	Combination	Quantity
1.	Baking Soda+ Distilled water	5 g BS + 50 ml DW
2.	Salt + Distilled water	5 g S + 50 ml DW
3.	Vinegar + Distilled water	4 ml Vi + 50 ml DW
4.	Baking soda + Vinegar + Distilled water	5 g BS + 4 ml Vi + 50 ml DW

5.	Salt + Vinegar + Distilled water	5 g S + 4 ml Vi + 50 ml DW
6.	Baking soda + salt + Vinegar + Distilled water	5 g BS + 5g S + 4 ml Vi + 50 ml DW

RESULTS AND DISCUSSION

In the present study it was found that *Pseudomonas aeruginosa* was found on all types of food garnishes whereas *E. coli* was found on Spring onion, Curry leaves and Black pepper. Similarly, *S. aureus* was found on Curry leaves and Black pepper while *Salmonella* sp. was found on Black pepper and Kasuri methi. *Citrobacter* sp. was found on Coriander and *Klebsiella* sp. was found on Black pepper (Table 5).

When antibiotic resistance profile of isolated bacteria was studied against 6 different antibiotics, it was found that all 100% bacteria were sensitive to Tetracycline, Levofloxacin and Gatifloxacin whereas 70% bacteria were resistant to Erythromycin. Followed by 30% bacteria are resistant to Azithromycin and 20% to Chloramphenicol (Table 6). According to Arslan et al (2011), the bacteria isolated from food garnishes were resistant to one or more antibiotics. That study reported that antibiotic resistance profile of all isolated bacteria were sensitive to Tetracycline, Levofloxacin and Gatifloxacin, while some are resistant as well as sensitive to Erithromycin, Azithromycin and Chloramphenicol.

The combination of Baking soda and Distilled water was found to be effective against 60% of isolated bacteria. The combination of Salt and Distilled water was found to be effective against 65% of isolated bacteria. The combination of Vinegar and Distilled water was found to be effective against 80% of isolated bacteria. The combination of Baking soda, Vinegar and Distilled water was found to be effective against 70% of isolated bacteria. The combination of Salt, Vinegar and Distilled water was found to be effective against 60% of isolated bacteria and the combination of Baking soda, Salt, Vinegar and Distilled water was found to be effective against 70% of isolated bacteria (Table 7). The combination of Vinegar and Distilled water were found to be the most effective decontaminant for bacteria isolated from food garnishes used in this study. This was the attempt to reduce chances of contaminantion of food garnishes followed in the study of (Banerjee and Sarkar, 2004).

Table No 5: Bacterial contamination on food garnishes

Sr. No.	Samples	Name of isolated bacteria
1.	Coriander (CO)	*Citrobacter* sp., *Pseudomonas aeruginosa*
2.	Springs onion (SP)	*E.coli* , *Pseudomonas aeruginosa*
3.	Curry leaves (CL)	*E. coli*, *Pseudomonas aeruginosa, S. aureus*
4.	Chilli papers (CP)	*Pseudomonas aeruginosa*
5.	Black paper (BP)	*E. coli*, *Pseudomonas aeruginosa, S. aureus* , *Salmonella* sp., *Klebsiella* sp.
6.	Kasuri methi (KM)	*Pseudomonas aeruginosa, Salmonella* sp.

Table No 6: Antibiotic Sensitivity profile of Bacteria isolated from food garnishes

Where, R= Resistant

Sr. No.	Isolated bacteria	Antibiotics					
		E	TE	LE	GAT	AZM	C
1.	*Citrobacter* sp.	19mm	18mm	26mm	27mm	15mm	21mm
2.	*Pseudomonas aeruginosa*	R	16mm	21mm	22mm	15mm	22mm
3.	*E. coli*	R	14mm	25mm	28mm	16mm	18mm
4.	*Pseudomonas aeruginosa*	R	17mm	29mm	30mm	21mm	23mm
5.	*E. coli*	23mm	21mm	26mm	24mm	R	24mm
6.	*S. aureus*	R	17mm	24mm	26mm	15mm	16mm
7.	*Pseudomonas aeruginosa*	R	12mm	29mm	28mm	22mm	26mm
8.	*Pseudomonas aeruginosa*	R	16mm	30mm	32mm	19mm	25mm
9.	*E. coli*	23mm	21mm	25mm	28mm	22mm	25mm
10.	*Pseudomonas aeruginosa*	R	17mm	30mm	32mm	20mm	24mm
11.	*S. aureus*	R	19mm	27mm	27mm	R	R
12.	*Salmonella* sp.	R	18mm	29mm	28mm	18mm	25mm
13.	*Klebsiella* sp.	R	19mm	30mm	31mm	R	R
14.	*Pseudomonas aeruginosa*	R	18mm	30mm	26mm	20mm	21mm
15.	*Salmonella* sp.	R	19mm	26mm	26mm	14mm	23mm

Table 7: Efficacy of Decontaminants on isolated bacteria from food garnishes

Where, S- Sensitive, R- Resistant

Sr. No.	Isolated bacteria	Baking soda+ Distilled water		Salt+ Distilled water		Vinegar+ Distilled water		Baking soda +vinegar+ Distilled water		Salt+Vinegar+ Distilled water		Baking soda+Salt+ Vinegar+ Distilled water	
1.	*Citrobacter* sp.(CO)	R		R		R		R		R		R	
2.	*E. coli*(SP)	R		S		R		S		R		S	
3.	*E. coli*(CL)	S		S		S		S		S		S	
4.	*E. coli*(BP)	S		S		R		S		S		S	
5.	*Klebsiella* sp.(BP)	S		S		R		R		S		R	
6.	*Salmonella* sp.(BP)	S		R		R		R		S		R	
7.	*Salmonella* sp.(KM)	R		R		R		R		R		R	
8.	*S. aureus*(CL)	R		R		R		R		R		R	
9.	*S. aureus*(CP)	S		R		R		R		S		R	
10.	*Pseudomonas aeruginosa*(CO)	R		R		R		R		R		R	
11.	*Pseudomonas aeruginosa*(SP)	R		R		R		R		R		R	
12.	*Pseudomonas aeruginosa*(CL)	R		R		R		R		R		R	
13.	*Pseudomonas aeruginosa*(CP)	R		R		R		R		R		R	
14.	*Pseudomonas aeruginosa*(BP)	S		S		S		S		S		S	
15.	*Pseudomonas aeruginosa*(KM)	R		R		R		R		R		R	
	Total	R	S	R	S	R	S	R	S	R	S	R	S
		9	6	10	5	13	2	11	4	9	6	11	4

CONCLUSION

In the present study, it is concluded that the most dominant bacteria on food garnishes was *Pseudomonas aeruginosa*. The decontaminants that were used were highly effective against bacteria which were isolated from different food garnishes that they have ability to destroy bacteria which were present on food garnishes. As food garnishes are used in uncooked form, so the chance of contamination and infection is very high. In this present study, it was concluded that the combination of vinegar and salt was found to be more effective against contaminant bacteria isolated from food garnishes. It helps to reduce the number of diseases that are caused by consumption of uncooked or unwashed garnishes.

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