

Assessment of the Bacteriological Quality of Food Samples May Offer Clues the Antibiotic Resistance Pattern

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Background: Foodborne illnesses continue to be a leading cause of morbidity and mortality worldwide; however, the burden of diseases caused by food-borne pathogens remains largely unknown.

Objectives: The aim of the present study was to culture-confirmed the bacterial profile and their antibiotic resistant pattern in Food and Drug Laboratory, Alborz University of Medical Sciences, Karaj, Iran.

Patients and Methods: A total of 22 bacteria including of *Staphylococcus aureus*, *Klebsiella spp* and *E. coli* were presumptive isolated from the traditional ice cream, cream pastries, sausage, and salami by the Official Food Microbiology Laboratory, Deputy of Food and Drug Administration, Alborz University of Medical Sciences, Karaj, Iran, and sent to the Research Center Laboratory, Alborz University of Medical Sciences, to confirm the bacterial spp by multiplex polymerase chain reaction. These isolates were also checked for their antimicrobial resistance pattern according to CLSI guideline.

Results: The highest rate of contamination was with *Klebsiella spp* 09 (40.9%), followed by *S. aureus* 07 (31.8%), *E. coli* 06 (27.27%), as reported by the Official Food Microbiology Laboratory of Alborz University of Medical Sciences. Gel electrophoresis of the isolates shows the 600bp bp and 80 bp gene among *S. aureus* and *E. coli* respectively. The antibiotic resistant pattern in case of *Klebsiella spp* showed that 6 (66.6%) *Klebsiella spp* were resistant to Penicillin and Cotrimoxazole. Similarly, penicillin and amoxicillin were found the highest resistant antibiotic against 83.3% *E. coli*, however, ceftriaxone showed the highest sensitivity against 100% *E. coli* isolates.

Conclusions: In conclusion, *Klebsiella spp*, *S. aureus* and *E. coli* are contaminants of food specimens obtained from food industries in Karaj, Iran; they constitute a serious health risk for human population. Moreover, the principal purpose of this study is to increase awareness of the antibiotic resistance of these bacteria poses threat.

Keywords: Antibiotic Resistance; *Klebsiella*; *Staphylococcus aureus*; *E. coli*

1. Background

The food industry is one of the fastest growing sections in the universal food Market. With the growing demand for food products worldwide, it is critical for manufacturers to take concerted actions to improve product quality and ensure product safety (1). The incidence of pathogenic bacteria has always been depended to factors like food, water, surfaces, equipments and most significantly food handlers (2). Improper handling of food is responsible for many food born diseases and intoxication (3, 4). Variety of bacterial species are found in a broad range of foods, including meat products, poultry and egg products, salads, bakery products, sandwich, milk and dairy products (5, 6). Many of these bacterial species can produce toxin which may cause any distur-

bance in the gastrointestinal tract (7, 8); thus an understanding of microbial profile of food is vital.

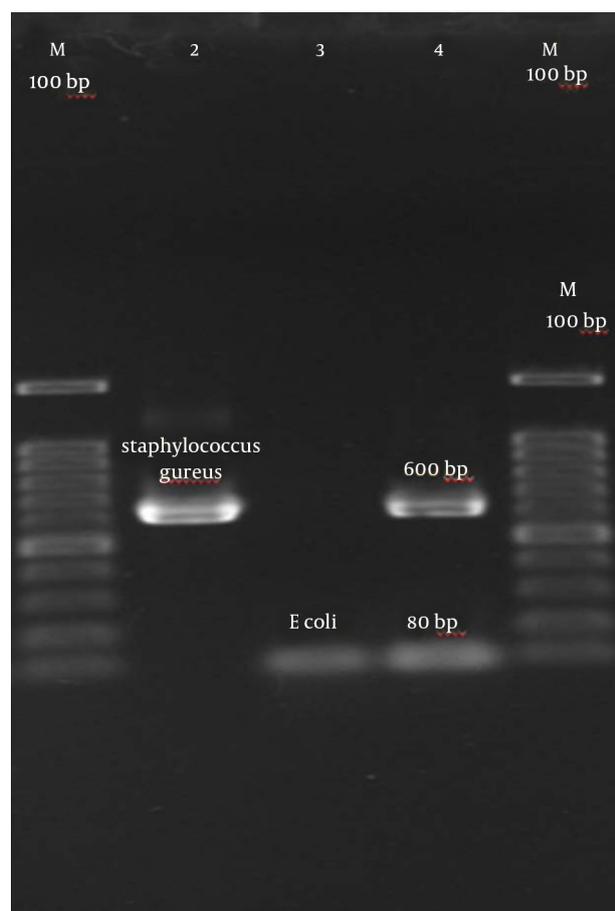
Above this, the emergence of antibiotic resistant food-borne pathogens is another public health concern. Several organizations, including the World Health Organization and Centers for Disease and Prevention (CDC), have reported the need to control the spread of this resistance (9).

2. Objectives

The aim of the present study was to culture-confirmed the bacterial profile and their antibiotic resistant pattern in Food and Drug Laboratory, Alborz University of Medical Sciences, Karaj, Iran.

Table 1. Most Expressive Antimicrobial Resistance Patterns of Isolated Bacteria From the Food Samples

Antibiotics	Resistance, %		
	<i>S. aureus</i> 7	<i>Klebsiella spp</i> 9	<i>E. coli</i> 6
Tetracycline	28.5	33.3	50
Chloramphenicol	14.2	ND	ND
Erythromycin	14.2	ND	ND
Gentamicin	0.0	22.2	33.3
Nitrofurantoin	14.2	11.1	16.6
Ceftriaxone	42.8	0.0	0.0
Amoxicillin	57.1	44.4	83.3
Amikacin	57.1	44.4	33.3
Penicillin	85.7	66.6	83.3
Co-trimoxazole	ND	66.6	66.6

Figure 1. Gel Electrophoresis of Identification of the 600 bp and 80 bp gene Among *S. aureus* and *E. coli*

Electrophoresis of PCR products introduced for 600 bp fragment of *Staphylococcus aureus* in 2nd well, 80 bp fragment of *E. coli* in 3rd well and multiplex PCR of *S. aureus* and *E. coli* in 4th well. The first well and the last well is 100 bp ladder (M).

3. Patients and Methods

A total of 22 bacteria including of *Staphylococcus aureus*, *Klebsiella spp*, and *E. coli* were presumptive isolated from the traditional ice cream, cream pastries, sausage, and salami by the Official Food Microbiology Laboratory, Deputy of Food and Drug Administration, Alborz University of Medical Sciences, Karaj, Iran, and sent to the Research Center Laboratory, Alborz University of Medical Sciences, to confirm the bacterial spp by multiplex polymerase chain reaction. Total bacterial DNA was extracted from pure cultured strains using kit according to manufacturer's instructions. These isolates were also checked for their antimicrobial resistance pattern according to CLSI guideline (10).

4. Results

The highest rate of contamination was with *Klebsiella spp* 09 (40.9%), followed by *S. aureus* 07 (31.8%), *E. coli* 06 (27.27%), as reported by the Official Food Microbiology Laboratory of Alborz University of Medical Sciences. To confirm the identification of these bacteria multiplex polymerase chain reaction could be done. Figure 1 shows gel electrophoresis of identification of the 600 bp and 80 bp gene among *S. aureus* and *E. coli* respectively.

Table 1 shows the antibiotic resistant pattern of the bacterial isolates. Overall, in case of *Klebsiella spp*, Penicillin and Cotrimoxazole was found the most resistant antibiotic as 6 (66.6%) *Klebsiella spp* were found resistant to these antibiotics. Similarly, penicillin and amoxicillin were found the highest resistant antibiotic against 83.3% *E. coli*, however, ceftriaxone showed the highest sensitivity against 100% *E. coli* isolates.

5. Discussion

Usually, conventional method is the standard procedure for the isolation and identification of many bacterial pathogens. However, this conventional method has many disadvantages; for example, we require a variety of enrichment broths and solid media to screen for all possible foodborne pathogens, and moreover it is time-consuming (11). To confirm the identification of isolated bacteria, we perform a multiplex PCR for the three types of bacteria isolated by Food Microbiology Laboratory, Deputy of Food and Drug, Alborz University of Medical Sciences. According to them the highest rate of contamination was by *Klebsiella spp* 09 (40.9%), followed by *S. aureus* 07 (31.8%), and *E. coli* 06 (27.27%). Various reports from all over the world revealed that bacterial foodborne agents have been the most well investigated and monitored causes of intestinal infectious disease and food-borne bacteria are increasingly becoming resistant to antimicrobials (12-14).

Among these bacteria, *Staphylococcus spp* have been known to cause *Staphylococcal* food poisoning due to ingestion of improperly stored food in which *Staphylococcus aureus* has grown. Our results revealed that locally

produced ice-cream were the most contaminated. Similar findings were reported by Mokhtarian et al. in Iran (15). Like other studies (16, 17), our study also revealed that *Escherichia coli* and *Klebsiella sp.* are capable of causing food borne diseases because we have also isolated these bacteria from the food samples taken by the Food Microbiology Laboratory, Deputy of Food and Drug, Alborz University of Medical Sciences, during their inspections.

The isolated and identified bacteria could be confirmed by using multiplex PCR carried out in Research laboratory, School of Medicine, Alborz University of Medical Sciences. Many scientists from all over the world reported the role and importance of multiplex PCR for detection and identification of variety of bacteria from food samples (17, 18).

It is worth to note that these bacteria showed a significant resistant pattern to a variety of antibiotics particularly penicillin. This finding is also similar to those reported by others (15, 16, 19, 20). Some scientists reported that presence of antibiotics in food of animals origin might hasten resistance development in humans. Therefore, types of antibiotics being used may help government officials minimize the risks of developing resistance; so further studies in this regard are necessary.

In conclusion, *Klebsiella spp.*, *S. aureus* and *E. coli* are contaminants of food specimens obtained from food industries in Karaj, Iran; they constitute a serious health risk for human population, and there is therefore a need for specific control programmes. Moreover, the principal purpose of this study is to increase awareness of the antibiotic resistance poses threat.

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Authors' Contributions

Enayatollah Kalantar desing the experiments and written the manuscript draft, Jafar Soltani scientific consultant and help in the first draft of the manuscript, Shiva Hatami culture confirm the isolates and antibiotic resistance experiments, Nooshin Habibi primary isolation of the bacteria, Helen Pomazaheri molecular experiments, Seyed Mahmoud Amin Marashi helped to designed the primers, Khatereh Amini help in sample collection, Afshin Afshar collection and management of sending the isolates, Kourosh Kabir analysed the data, Ali Akabr Safari help in designed the experiments.

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