Prevalence of *Staphylococcus aureus* in Shrimps in Tehran during 2013

Mohammad Mehdi Soltan Dallal 1, 2, Abbas Rahimi Foroushani 3, Sara Sharifi –Yazdi 4, Mohammad Kazem Sharifi -Yazdi 5, 6*, Noushin Arfatahery 2

1 Food Microbiology Research Center, Tehran University of Medical Sciences, Tehran, IR Iran.

2 Division of Food Microbiology, Department of Pathobiology, School of Public Health, Tehran University of Medical Sciences. Tehran, IR Iran.

3 Department of Epidemiology and Biostatics, School of Public Health, Tehran University of Medical Sciences Tehran, IR Iran.

4 Medical Student, School of Medicine, Tehran University of Medical Sciences, Tehran, IR Iran.

5 Zoonosis Research Centre, Tehran University of Medical Sciences, Tehran, IR Iran.

6 Department of Medical Laboratory Sciences, School of Paramedicine, Tehran University of Medical Sciences. Tehran, IR Iran.

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**ABSTRACT**

**Background**
During fishing and transport, preservation and quality of fish products are important as well as storage to prevent the growth of pathogenic and toxin producing bacteria. *Staphylococcus aureus* is one of the most common causes of sea food-borne diseases worldwide due to contamination of food by preformed enterotoxins. The aim of this study was to compare the prevalence and contamination of *S. aureus* in marine and farmed shrimps in Tehran fishery center.

**Methods:** A total of 300 samples, including 150 marine, 150 farmed shrimps were selected during September 2013 to December 2014. Isolation and identification of *S. aureus* from isolated samples were carried out according to conventional methods, and antibiotic susceptibility test was performed by modified Kirby-Bauer disc diffusion method.

**Results:** The results of this study showed that 30% of marine and 20% off armed shrimps were contaminated with *S. aureus*. The highest resistance was observed with penicillin and ampicillin, whereas 100% were sensitive to vancomycin, clindamycin, ciprofloxacin, and rifampin.

**Conclusions:** Due to relatively high contamination of shrimp by *S. aureus* more attention should be given during processing and manufacturing.

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* Corresponding Author: Mohammad Kazem Sharifi -Yazdi, Department of Medical Laboratory Sciences, School of Para Medicine, Tehran University of Medical Sciences. Tehran, IR Iran. Tel: +98-21-8893919, Email: mksharifi@tums.ac.ir
Introduction

Seafood is the most essential nutritional needs of each community. Often, consumption of contaminated fish and shrimp could cause gastrointestinal diseases in human (1). Improper cooking of these products, which are often cooked or grilled or fried, usually results in the center of meat of being raw and pathogens or heat-stable toxins could remain unchanged which can cause illness. Staphylococcus aureus, toxins and heat-resistant enterotoxins, SEA, SEB, can cause gastroenteritis, gastrointestinal toxicity, fever, nausea, dysentery and colitis (2, 3). Standard procedures and preserve the quality of these products are important during fishing, transportation, and storage to prevent the growth of pathogenic bacteria and toxin production. The potential transmission of this dangerous strain of human carriers or the environment, such as transport and packaging, contaminated hands of workers or infected respiratory secretions to seafood products has been studied in other countries (4, 5). In some parts of the world, more than 50% of food poisoning is caused by SEA. In Great Britain and America, SEA and SEB cause more than 69 percent of food poisoning (3, 5). Toxic shock syndrome toxin (TSST) causes systemic infection, shock and anaphylactoid reactions, inpatients and pregnant women through the gastrointestinal absorption and blood transfusion (6, 7). The children, pregnant women, the elderly and patients who are undergoing chemotherapy or taking immune suppressing drugs would be more severe and the health status of these individuals who are consuming shrimps could be in greater danger (8). On the other hand, because of its texture, fish and shrimp meat have a high plasticity of corruption. Improper conditions in the fishery, storage and non-standard transportation provide conditions for pathogens growth. Unfortunately, improper cooking of these products is the most important reason in causing diseases.

All these factor shoveled to the increased risk of gastroenteritis and food poisoning caused by contaminated food. Food borne diseases are a large group of the world diseases and are one of the most important problems in each community (9). For instance in America, food borne diseases are at the third level after heart disease and respiratory (10, 11). According to national health agencies, average food illness outbreak in the EU and third world countries have reached, 38.3 and 91.58 cases per 100000 populations (12, 13). The Food Standards Agency has legislated guidelines for fresh and frozen fish and shrimp microbial control (14, 15).

Material and method

Samples

A total of 300 samples, including 150 marine, royal shrimps and 150 farmed shrimps (L. vannamei) with the healthy appearance were selected during September 2013 to December 2014. One gram of shrimp meat was cut with the sterile scalpel, mixed with guilt. All samples were brought to food microbiology laboratory, School of Public Health, Tehran University of Medical Sciences, in sterile container containing ice quickly and were stored in a refrigerator at 4 °C until tested. Three experiments were done on each shrimp, involved its head, meat and the first part of tail. The experiments were conducted according to food standard No. 2325 (14) and Iran national standard No. 356 in sterile condition.

Identification

One gram of shrimp meat was cut with sterile scalpel, mixed with guilt, and the solution was brought into suspension, according to Iran National Standard protocol No. 6803 (15). S. aureus ATCC 29213 and E. coli ATCC 25922 were used as positive and negative controls. Tubes were incubated at 37 °C for 24-48 hrs. After the time, according to standard No. 2747, the tubes that had
deposition or were black, were cultured in Baird–Parker agar with 0.1% potassium tellurite solution and egg emulsion with surface culture method and incubated at 37 °C for 24-48 hrs.

Small colonies of black, shiny with aid of oil zone microscope, indicating *S. aureus* were selected. Coagulase test was done and the tubes containing clot was confirmed as coagulase-positive *S. aureus*.

**Antimicrobial Susceptibility Testing**

Susceptibilities of 75 strains of *S. aureus* were determined against 12 antimicrobial drugs using the disk agar diffusion method. The isolates were examined for their susceptibilities to ampicillin, chloramphenicol, gentamicin, tetracycline, ciprofloxacin, penicillin, erythromycin, clindamycin, oxacillin, rifampin and vancomycin (Mast, UK) by the standard disc diffusion method (16).

**Statistical Analysis**

Statistical analysis was performed with SPSS/PC 11.5 software (SPSS, Chicago, IL). The chi-square test and Fisher's exact two-tailed test were used for statistical analysis. A P value less than 0.05 was considered statistically significant.

**Result**

As it is shown in table 1, from the total of 300, 75 tested samples were contaminated.

**Table 1.** Contamination rate of marine and farmed shrimps.

<table>
<thead>
<tr>
<th>samples</th>
<th>Number of samples</th>
<th>Number of samples contaminated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Marine shrimps</td>
<td>150</td>
<td>45 (30%)</td>
</tr>
<tr>
<td>Farmed shrimps</td>
<td>150</td>
<td>30 (20%)</td>
</tr>
</tbody>
</table>

The marine shrimps were more contaminated, 45 (30%), than farmed shrimps 30 (20%).

Table 2 shows the antimicrobial susceptibility patterns of *Staphylococcus aureus*. The highest sensitivity (100%) was observed with to clindamycin, rifampin, ciprofloxacin and vancomycin, whereas approximately (76 %) were resistance to ampicillin and penicillin.

**Table 2.** Antimicrobial resistance patterns of *S. aureus* isolates.

<table>
<thead>
<tr>
<th>Antibiotic</th>
<th>Sensitive No (%)</th>
<th>Resistance No (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Penicillin</td>
<td>18 (24)</td>
<td>57 (76)</td>
</tr>
<tr>
<td>Ampicillin</td>
<td>17 (22.7)</td>
<td>58 (77.3)</td>
</tr>
<tr>
<td>Oxacillin</td>
<td>52 (69.3)</td>
<td>23 (30.7)</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>72 (96)</td>
<td>3 (4)</td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>75 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Erythromycin</td>
<td>69 (92)</td>
<td>6 (8)</td>
</tr>
<tr>
<td>Clindamycin</td>
<td>75 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Vancomycin</td>
<td>75 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>73 (97.3)</td>
<td>2 (2.7)</td>
</tr>
<tr>
<td>Tetracycline</td>
<td>59 (78.7)</td>
<td>16 (21.3)</td>
</tr>
<tr>
<td>Rifampin</td>
<td>75 (100)</td>
<td>0 (0)</td>
</tr>
</tbody>
</table>
Discussion

Food safety plays an important role in community health. Despite continuing advances in knowledge and techniques in food safety, consumption of contaminated food is still direct cause of many diseases. A study conducted in Iceland showed that out of total of 7913 freshly shrimps only 2% were contaminated with S. aureus (17). Their results were contradicted with the results of our study. This might be due to low water temperature in Iceland or better handling during manufacturing and distribution. In a similar study carried out in India the rate of contamination with S. aureus was 5% which was much lower than our finding (18). Apart from one study that reported high rate of contamination with S. aureus (19, 20), most of other researcher reported lower rate of S. aureus than our results (21, 22). In our study marine shrimp was more contaminated (30%) than farmed shrimp (20%) which might be due to better handling of farmed shrimp. The susceptibility testing of isolates revealed higher resistance towards ampicillin (77.3%) and penicillin (76%) and no resistance towards rifampin, vancomycin, clindamycin and ciprofloxacin (100% each), indicating that these antibiotics might be an option for empirical therapy.

Conclusion

High rate of contamination of shrimp obtained in this study showed that urgent attention needs to prevalent S. aureus among aqua products. The information provided here can be used as a useful database for epidemiological purposes, although more study is needed to verify these results.

Acknowledgements

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Conflict of interest

None declared conflicts of interest.

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