The study of bacterial pathogens, *Staphylococcus aureus*, *Vibrio parahaemolyticus* and *Escherichia coli*, in fresh and smoked cultivated fish in Iran

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There is an increasing demand for fish and fish products around the world (Feldhusen 2004). However, there is substantial evidence that fish and seafood are high on the list of foods associated with outbreaks of food borne diseases (Huss 2004). A large proportion of these outbreaks are caused by biotoxins, histamines and viruses. Nevertheless, fish and seafood also may be a vehicle for many bacterial pathogens (Davis et al. 2003, Hosseini et al. 2004).

In assessing the risks from fish, it is important to have information on the incidence of these pathogens (Davis et al. 2003). The microbial status of seafood after catch is closely related to environmental conditions and the microbiological quality of the water. These factors include water temperature, salt content, distance between localization of catch and polluted areas, especially those containing human and animal feces, natural occurrence of bacteria in the water, ingestion of food by fish, methods of catch and chilling, and postharvest handling or processing conditions (Feldhusen 2004).

Salted and smoked fish are traditional products in Iran that are prepared by heavy salting followed by a cold smoking method in northern Iran (Gilan and Mazandaran provinces). These products are prepared from fresh or cultured fish caught from fish ponds that are commonly fertilized with cow manure or the Caspian Sea. The smoked fish are usually kept in plastic bags outside the refrigerator in fish markets. Salted and smoked fish are traditionally consumed raw or after exposure to inadequate cooking temperatures (Nikpay 2004). Therefore, consumption of them may lead to food borne diseases. In this study, the incidence of some bacterial pathogens (*Staphylococcus aureus*, *Vibrio parahaemolyticus* and *Escherichia coli*) in fresh and smoked fish were investigated.

**Study Methods**

One hundred fish were sampled. Fifty anadromous shad (*Alosa kessleri*) were caught in the Caspian Sea near the coast and and 50 cultured silver carp were obtained from a fish farm in Mazandaran. The fish were sampled aseptically and bacterial testing followed the American Public Health Association and U.S Food and Drug Administration isolation methods (APHA 1997, FDA 2004). Twenty-five wild fish and 25 cultured fish samples were eviscerated and traditionally salted (30-35 kg salt for 100 kg fish) in a watertight container and were smoked using a cold smoking method at temperatures not exceeding 32°C for one week. All of these processes were undertaken according to the practices used in a commercial processing facility in Mazandaran province. The smoked fish were tested for various pathogenic bacteria at the end of the smoking process. The APHA method for isolation of *S. aureus* including enrichment of a 1g sample in 10 mL cooked meat media plus 9 percent NaCl (W/v), streaking a loopful of the 24h enrichment culture on Baird-Parker agar containing egg yolk agar and potassium tellurite and subsequent confirmatory coagulase test of lipase-positive jet-black colonies (AOAC 1995). The APHA method for isolation of *V. parahaemolyticus* was as follows: 25 g of sample was homogenized in 225 mL alkaline peptone water, using a stomacher and incubated at 35°C for 6-8 h. A loopful from APW was then streaked onto thiosulfate citrate bile salt sucrose agar (TCBS) and the plate was incubated at 35°C for 24h. Confirmation tests were then made on blue-green colonies for acid production from cellobiose, sucrose, maltose, mannitol, trehalose and lactose, Gram staining, motility, Vogues-proskauer, oxidase and nitrite reduction tests, growth in 6 and 8 percent NaCl and growth at 43°C (APHA 1997).

We also used the APHA methods for detection of *E. coli* in salted and smoked fresh and cultivated fish. For coliform, violet red bile agar (VRBA ) was used. The pour plate technique was applied for the coliform count, while for *S. aureus* plate count, surface plate was used. To confirm the suspected coliforms, 10 of the purple-red colonies, 0.5 mm in diameter or larger, surrounded by a zone of precipitated bile acids on VRBA were transferred to tubes of brilliant-green lactose bile broth at 35°C for 24-48h. The confirmed coliform colonies were also assessed for detection of *E. coli*, using Eosin-methylene-blue lactose and serological tests (APHA1997).
In conclusion, this study indicates that S. aureus and V. parahaemolyticus may be present in salted, cold-smoking fish. Because these products are consumed raw or after exposure to inadequate cooking temperatures in Iran, consumption of them may pose a risk of food borne infections and intoxication to consumers.

Notes

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References


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